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(54) Title: ION MOBILITY AND MASS SPECTROMETER

(57) Abstract

An ion mobility and mass spectrometer instrument (300) includes an ion source region (32) coupled to an ion mobility spectrometer (34) having an ion outlet coupled to a quadrupole mass filter (302). An output of the filter is coupled to a collision cell (304) which has an ion outlet coupled to an ion acceleration region of a mass spectrometer (36) such as a time of flight mass spectrometer. The instrument (300) is particularly well suited for sequencing analysis wherein a sample is ionized and a resulting three-dimensional ion spectrum (ion intensity vs. ion mobility and ion mass) is observed. If the spectrum reveals that no ions overlap in mobility values, the collision cell (304) is filled with a suitable buffer gas (306) and the instrument (300) is reactivated whereby a complete three-dimensional spectrum of parent and daughter ions results. If, however, the original spectrum reveals that two or more ions overlap in ion mobility values, the collision cell (304) is filled with a buffer gas (306) and the quadrupole mass filter (302) is controlled to selectively filter out all but one of the ions having overlapping mobility values. The instrument (300) is reactivated, and the quadrupole mass filter (302) is selectively controlled, as many times as mobility overlap occurs to thereby provide complete three-dimensional spectra of parent and daughter ions resulting from fragmentation. Various configurations of mass filter (302), ion trap (32) and collision cell (304) positioning, relative to the ion mobility (34) and mass spectrometer (36) instruments, are contemplated.

ION MOBILITY AND MASS SPECTROMETER

CROSS-REFERENCE TO RELATED U.S.APPLICATION

This is a continuation-in-part of co-pending U.S.
5   Patent Application Ser. No. 08/867,245, filed June 2,
1997 and entitled HYBRID ION MOBILITY AND MASS
SPECTROMETER.

Field of the Invention

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The present invention relates generally to
instrumentation for characterization of molecules based
on their structures and mass-to-charge ratios as gas-
phase ions, and more specifically to such
15   instrumentation which provides for rapid and sensitive
analysis of composition, sequence, and/or structural
information relating to organic molecules, including
biomolecules, and inorganic molecules.

BACKGROUND OF THE INVENTION

20

Biological molecules, such as DNA, RNA, proteins,
carbohydrates and glycoconjugates, are comprised of
repeating subunits typically referred to as residues.
25   The sequence of such residues ultimately defines the
structure and function of the biomolecule and determines
how it will interact with other molecules.

A central part of almost all conventional
sequencing strategies is the analysis of complex sets of
30   sequence-related molecular fragments by chromatography
or by polyacrylamide gel electrophoresis (PAGE). PAGE-

based automated sequencing instruments currently exist and typically require a number of fluorescent dyes to be incorporated into the base-specifically terminated biomolecule product, which is then processed through the
5 polyacrylamide gel. The discrete-length product molecules are detected near the bottom of the gel by their emitted fluorescence following excitation by a radiation source.

Such automated instruments are typically capable of
10 generating sequence information for biomolecules having 500 or more residues at a rate of 10-20 times faster than manual methods. However, both the manual and automated PAGE techniques suffer from several drawbacks. For example, both approaches are labor-intensive since a
15 gel must be prepared for each sequencing run. Also, while automated PAGE systems may offer faster analysis times than a manual approach, the accuracy of such systems is limited by artifacts generated by non-uniform gel matrices and other factors. Such automated systems
20 are generally not equipped to accurately process the effects of such artifacts, which are typically manifested as "smiling" compressions, faint ghost bands, and the like. Manual interpretation of such results is therefore often required which significantly increases
25 analysis time.

Researchers have, within the past several years, recognized a need for more rapid and sensitive techniques for analyzing the structure and sequences of biomolecules. Mass spectrometry (MS) techniques, such
30 as time-of-flight mass spectrometry (TOFMS) and Fourier Transform ion-cyclotron-resonance mass spectroscopy, are

well known techniques for quickly and accurately providing ion mass information from which sequence and structural determinations can be made. As is known in the art, TOFMS systems accelerate ions, via an electric

5   field, toward a field-free flight tube which terminates at an ion detector. In accordance with known TOFMS principles, ion flight time is a function of ion mass so that ions having less mass arrive at the detector more quickly than those having greater mass. Ion mass can

10   thus be computed from ion flight time through the instrument. FIG. 1 demonstrates this principle for a cytochrome-c sample, having a known mass to charge ratio (m/z) of 12,360 da, and a lysozyme sample, having a known mass to charge ratio (m/z) of 14,306 da. In FIG.

15   1, signal peak 10, having a flight time of approximately 40.52 μs corresponds to the lighter cytochrome-c sample, and signal peak 12, having a flight time of approximately 41.04 μs, corresponds to the heavier lysozyme sample.

20   Due to the significantly decreased sample preparation and analysis times of MS techniques over the above-described PAGE technique, several MS sequencing strategies have recently been developed. Such MS sequencing techniques are generally operable to measure

25   the change in mass of a biomolecule as residues are sequentially removed from its end. Examples of two such techniques, each involving elaborate pre-MS processing techniques, are described in U.S. Patent Nos. 5,210,412 to Levis et al. and 5,622,824 to Köster.

In order to provide for the capability of
determining sequence and structural information for
large biomolecules, it has been recognized that MS
techniques must accordingly be capable of generating
5   large ions. Currently, at least two techniques are
known for generating large ions for spectral analysis;
namely electrospray ionization (ESI) and matrix assisted
laser desorption ionization (MALDI). While both large
ion generating techniques are readily available, known
10   MS techniques are limited in both the quantity and
quality of discernable information. Specifically, for
large biomolecules, defined here as those containing at
least 50 residues, mass spectra of parent and sequence
related fragment ions become congested to the degree
15   that mass (TOF) peaks overlap.

One solution to the problem of congested mass
spectra is to increase the mass resolution capability of
the MS instrument. Recent efforts at increasing such
resolution have been successful, and complete sequence
20   information for a 50 base pair DNA has been obtained
using a Fourier Transform ion cyclotron resonance
(FTICR) instrument. However, such instruments are
extremely expensive, not readily available, and because
of their extremely high vacuum requirements, they are
25   generally not suitable for routinely sequencing large
numbers of samples.

Another solution to the problem of congested mass
spectra is to pre-separate the bulk of ions in time
prior to supplying them to the ion acceleration region
30   of the MS instrument. Mass spectrometry can then be
performed sequentially on "packets" of separated ion

samples, rather than simultaneously on the bulk of the
generated ions. In this manner, mass spectral
information provided by the MS instrument may be spread
out in another dimension to thereby reduce the localized
5   congestion of mass information associated with the bulk
ion analysis.

One known ion separation technique which may be
used to pre-separate the bulk of the ions in time prior
to MS analysis is ion mobility spectrometry (IMS). As
10   is known in the art, IMS instruments typically include a
pressurized static buffer gas contained in a drift tube
which defines a constant electric field from one end of
the tube to the other. Gaseous ions entering the
constant electric field area are accelerated thereby and
15   experience repeated collisions with the buffer gas
molecules as they travel through the drift tube. As a
result of the repeated accelerations and collisions,
each of the gaseous ions achieves a constant velocity
through the drift tube. The ratio of ion velocity to
20   the magnitude of the electric field defines an ion
mobility, wherein the mobility of any given ion through
a high pressure buffer gas is a function of the
collision cross-section of the ion with the buffer gas
and the charge of the ion. Generally, compact
25   conformers, i.e. those having smaller collision cross-
sectional areas, have higher mobilities, and hence
higher velocities through the buffer gas, than diffuse
conformers of the same mass, i.e. those having larger
collision cross-sectional areas. Thus, ions having
30   larger collision cross-sections move more slowly through
the drift tube of an IMS instrument than those having

smaller collision cross-sections, even though the ions
having smaller collision cross-sections may have greater
mass than those having higher collision cross-sections.
This concept is illustrated in FIG. 2 which shows drift

5   times through a conventional IMS instrument for three
ions, each having a different mass and shape (collision
cross-section). As is evident from FIG. 2, the most
compact ion 14 (which appears to have the greatest mass)
has the shortest drift time peak 16 of approximately 5.0

10   ms, the most diffuse ion 18 has the longest drift time
peak 20 of approximately 7.4 ms, and the ion 22 having a
collision cross-section between that of ion 14 and ion
18 (which also appears to have the least mass), has a
drift time peak 24 of approximately 6.1 ms.

15   Referring now to FIG. 3, an ion time-of-flight
spectrum 26, obtained from a known time-of-flight mass
spectrometer, is shown plotted vs. ion drift time. In
this figure, ions of different mass are dispersed over
different times of flight in the mass spectrometer.

20   However, due to the limited resolution of the mass
spectrometer, ions are not completely separated in the
spectrum, i.e. dots corresponding to different ions
overlap. When compared with FIG. 6, which will be
discussed more fully in the DESCRIPTION OF THE PREFERRED

25   EMBODIMENTS section, it is evident that different ions
can be better resolved by an instrument that separates
ions in two dimensions, namely ion mobility and ion
mass.

             Guevremont et al. have recently modified an

30   existing IMS/MS instrument to convert a quadrupole MS to
a TOFMS [R. Guevremont, K.W.M. Siu, and L. Ding,

PROCEEDINGS OF THE 44TH ASMS CONFERENCE, (1996),
Abstract]. Ions are generated in the Guevremont et al.
instrument via electrospray, and 5 ms packets are gated
into the IMS instrument. The ion packets produced by
5   the IMS instrument are passed through a small opening
into an ion acceleration region of the TOFMS.

While Guevremont et al. have had some experimental
success in coupling an IMS instrument to a TOFMS
instrument, their resulting instrumentation and
10   techniques have several drawbacks associated therewith.
For example, since the Guevremont et al. abstract
discusses using 5ms gate pulses to admit ions into the
IMS instrument, it is noted that the resultant IMS
spectrum has low resolution with at least 5 ms peak
15   widths. Secondly, because the drift tube and ion flight
tube of the Guevremont et al. instrument are colinear,
any spatial and temporal spread in an ion packet leaving
the IMS leads directly to a spatial and temporal spread
of ions in the ion acceleration region of the TOFMS.
20   These two characteristics lead to poor mass resolution
in the TOFMS. The combination of low resolution in the
IMS and low resolution in the TOFMS makes this
instrument incapable of resolving complex mixtures.
What is therefore needed is a hybrid IMS/TOFMS
25   instrument optimized to resolve complex mixtures. Such
an instrument should ideally provide for optimization of
the ion mobility spectrum as well as optimization of the
mass spectrum. Moreover, such a system should provide
for an optimum interface between the two instruments to
30   thereby maximize the capabilities of the TOFMS.

SUMMARY OF THE INVENTION

The foregoing drawbacks associated with the prior art systems discussed in the BACKGROUND section are
5 addressed by the present invention. In accordance with one aspect of the present invention, a method of generating ion mass spectral information comprises the steps of generating a gaseous bulk of ions, gating at least a portion of the bulk of ions into an ion mobility
10 spectrometer to thereby separate the bulk of ions in time to form a number of ion packets each having an ion mobility associated therewith, sequentially directing at least some of the ion packets into a mass spectrometer, continually activating the mass spectrometer to thereby
15 sequentially separate at least some of the ion packets in time to form a number of ion subpackets each having an ion mass associated therewith, and processing at least some of the ion subpackets to determine mass spectral information therefrom.
20   In accordance with another aspect of the present invention, an apparatus for generating mass spectral information from a sample source comprises means for generating a gaseous bulk of ions from a sample source, an ion mobility spectrometer (IMS) having an ion inlet
25 coupled to the means for generating a gaseous bulk of ions and an ion outlet, wherein the IMS is operable to separate ions in time as a function of ion mobility, a mass spectrometer (MS) having an ion acceleration region coupled to said ion outlet of said IMS and an ion
30 detector, wherein the MS is operable to separate ions in time as a function of ion mass, and a computer operable

to gate at least a portion of the gaseous bulk of ions
into the ion inlet of the IMS and to continually pulse
the ion acceleration region of the MS to thereby
sequentially direct ions toward the ion detector.

5   In accordance with yet another aspect of the
present invention, a method of generating ion mass
spectral information comprises the steps of generating a
gaseous bulk of ions, separating the gaseous bulk of
ions in time as a function of ion mobility, where two or

10   more ions overlap in ion mobility values, filtering out
ions that have all but a desired mass-to-charge ratio,
sequentially separating in time the post-filtered ions
as a function of ion mass, and processing ions separated
as two-dimensional functions of ion mobility and ion

15   mass to determine ion mass spectral information
therefrom.

In accordance with a further aspect of the present
invention, an apparatus for generating mass spectral
information from a sample source comprises means for

20   generating a gaseous bulk of ions from a sample source,
an ion mobility spectrometer (IMS) having an ion inlet
coupled to the means for generating a gaseous bulk of
ions and an ion outlet, wherein the IMS is operable to
separate ions in time as a function of ion mobility, an

25   ion filter having a filter inlet coupled to the ion
outlet of the IMS and a filter outlet, wherein the ion
filter is operable to sequentially pass therethrough
only ions having desired mass-to-charge ratios, and a
mass spectrometer (MS) having an ion acceleration region

30   coupled to the filter outlet and an ion detector,
wherein the MS is operable to sequentially separate in

time ions provided thereto by the ion filter as a
function of ion mass.

In accordance with still a further aspect of the
present invention, a method of generating ion mass

5   spectral information comprises the steps of generating a
gaseous bulk of ions, separating the gaseous bulk of
ions in time as a function of ion mobility, sequentially
separating in time as a function of ion mass each of the
ions separated in time as a function of ion mobility,

10 processing ions separated as two-dimensional functions
of ion mobility and ion mass to determine ion mass
spectral information therefrom, repeating the generating
and separating steps followed by the step of
sequentially fragmenting into daughter ions each of the

15 ions separated in time as a function of ion mobility,
followed by the sequentially separating and processing
steps only if the initial processing step indicates that
no two or more ions overlap in mobility values. If, on
the other hand, the initial processing step indicates

20 that two or more ions overlap in mobility values, the
method further includes the step of filtering out ions
that have all but a desired mass-to-charge ratio,
followed by repeating the generating and separating
steps, followed by the step of sequentially fragmenting

25 into daughter ions each of the ions separated in time as
a function of ion mobility, followed by the sequentially
separating and processing steps. Thereafter, the method
further includes the step of repeating the filtering
step until all ions overlapping in ion mobility values

30 have been processed.

10

One object of the present invention is to provide instrumentation for rapid analysis and sequencing of large biomolecules, as well as analysis of mixtures of organic and inorganic molecules.

5   Another object of the present invention is to provide an ion mobility and time-of-flight spectrometer for composition, sequence and structural analysis of biomolecules.

Yet another object of the present invention is to

10   optimize such an instrument for sensitivity and resolution of both ion mobility and ion mass spectra.

Still another object of the present invention is to provide a technique for operating such an instrument in obtaining sequencing information.

15   These and other objects of the present invention will become more apparent from the following description of the preferred embodiments.

BRIEF DESCRIPTION OF THE DRAWINGS

FIG. 1 is a MALDI-TOF mass spectrum of cytochrome-c and lysozyme.

FIG. 2 is an IMS drift time distribution for three ions having different collision cross-sections.

FIG. 3 is a mass spectrum plotted against drift time illustrating the limited resolution of a time-of-flight mass spectrometer.

FIG. 4 is a cross-section and schematic diagram of one embodiment of a hybrid ion mobility and time-of-flight mass spectrometer, in accordance with the present invention.

FIG. 5 is a cross-section and schematic diagram of an alternate embodiment of a hybrid ion mobility and time-of-flight mass spectrometer, according to the present invention.

FIG. 6 is a plot of ion time-of-flight vs. ion drift time for oligothymidine, utilizing the hybrid instrumentation of either FIG. 4 or FIG. 5.

FIG. 7A is a diagrammatic illustration of one preferred embodiment of an ion source for use with any of the instrument configurations shown in FIGS. 4, 5 and 9.

FIG. 7B is a diagrammatic illustration of an alternate embodiment of an ion source for use with any of the instrument configurations shown in FIGS. 4, 5 and 9.

FIG. 7C is a diagrammatic illustration of another alternate embodiment of an ion source for use with any

of the instrument configurations shown in FIGS. 4, 5 and 9.

FIG. 8A is a plot of ion intensity vs. ion drift time for an IMS instrument without an ion trap disposed

5   between the ion source and the IMS instrument.

FIG. 8B is a plot of ion intensity vs. ion drift time for an IMS instrument having an ion trap disposed between the ion source and the IMS instrument.

FIG. 9 is a block diagram illustration of an

10   another alternate embodiment of an ion mobility and time-of-flight mass spectrometer, in accordance with the present invention.

FIG. 10 is a partial cross-sectional diagram of yet another alternate embodiment of an ion source for use

15   with any of the instrument configurations shown in FIGS. 4, 5 and 9.

FIG. 11 is a cross-section of one preferred embodiment of the quadrupole mass filter illustrated in FIG. 9 as viewed along section lines 11-11.

20   FIG. 12 is a plot of ion intensity vs. mass-to-charge ratio illustrating operation of the quadrupole mass filter of FIG. 11.

FIG. 13 is a flowchart illustrating one preferred embodiment of a process for conducting sequencing

25   analysis using the instrument configuration of FIG. 9, in accordance with the present invention.

FIG. 14 is composed of FIGS. 14A-14D and illustrates an example ion mass/mobility spectrum resulting from a first pass through the process

30   illustrated in FIG. 13.

FIG. 15 is composed of FIGS. 15A-15D and illustrates an example ion mass/mobility spectrum resulting from a second pass through the process illustrated in FIG. 13.

5   FIG. 16 is composed of FIGS. 16A-16D and illustrates an example ion mass/mobility spectrum resulting from a third pass through the process illustrated in FIG. 13.

FIG. 17 is a block diagram illustrating alternative
10 structural variations of the ion mobility and time-of-flight mass spectrometer of the present invention.

DESCRIPTION OF THE PREFERRED EMBODIMENTS

For the purposes of promoting an understanding of the principles of the invention, reference will now be made to the embodiments illustrated in the drawings and specific language will be used to describe the same. It will nevertheless be understood that no limitation of the scope of the invention is thereby intended, such alterations and further modifications in the illustrated devices, and such further applications of the principles of the invention as illustrated therein being contemplated as would normally occur to one skilled in the art to which the invention relates.

Referring now to FIG. 4, one preferred embodiment of a hybrid ion mobility and time-of-flight mass spectrometer instrument 30, in accordance with the present invention, is shown. Instrument 30 includes, as its basic components, an ion source region 32 in communication with an ion mobility spectrometer 34, which itself is in communication with a mass spectrometer 36. A computer 38 is provided for controlling at least some portions of the instrument 30 as well as for collecting ion information from mass spectrometer 36. Computer 38 is preferably a personal computer (PC) of known construction having at least a known 386 processor, although the present invention contemplates that computer 38 may be any known computer, controller or data processor capable of controlling instrument 30, as set forth in greater detail hereinafter, and of collecting and processing ion information from mass spectrometer 36.

Preferably, mass spectrometer 36 is of the linear
time-of-flight type, although the present invention
contemplates that spectrometer 36 may alternatively be a
known reflectron time-of-flight mass spectrometer,
5   multi-pass time-of-flight mass spectrometer or Fourier
Transform ion-cyclotron-resonance (FTICR-MS) mass
spectrometer. In one preferred embodiment, the TOFMS 36
is configured to maximize mass resolution by minimizing
the deleterious effects of initial ion position and
10   initial ion velocity distributions. Details of such a
TOFMS configuration and operation thereof are given in
U.S. Patent Nos. 5,504,326, 5,510,613 and 5,712,479 to
Reilly et al., all assigned to the assignee of the
present invention, and the contents of which are all
15   incorporated herein by reference.

Ion mobility spectrometer (IMS) 34 includes a drift
tube 40 having a gas port 42 disposed adjacent to an ion
exit end 44 of tube 40, wherein port 42 is connected to
a source of buffer gas 46. The flow rate of buffer gas
20   may be controlled by computer 38 via signal path 48, or
may alternatively be controlled by a manually actuated
valve (not shown). Ion exit end 44 of drift tube 40
includes an endplate 43 attached thereto, wherein
endplate 43 defines an opening, or ion aperture, 45
25   therethrough.

Drift tube 40 includes a number of guard rings 50
distributed along its inner surface, wherein the guard
rings 50 are interconnected by equivalent-valued
resistors (not shown). The guard ring positioned most
30   adjacent to ion source region 32 is connected to a
voltage source VS1 52 via signal path 54, and source 52

is preferably controlled by computer 38 via signal path
56, although the present invention contemplates
controlling source 52 via a manual actuator (not shown).
The drift tube 40 defines a longitudinal axis 72

5   therethrough which will be referred to hereinafter as
the drift tube axis 72. Voltage source 52 is preferably
set to a positive voltage to thereby establish a
constant electric field directed along axis 72 in a
direction indicated by arrow 55. Those skilled in the

10 art will recognize that the importance of the guard ring
and voltage source arrangement of the spectrometer 34
lies not in its specific structure, but in its ability
to establish, as accurately as possible, a constant
electric field in the direction of arrow 55. In this

15 sense, the present invention contemplates that any known
structure or arrangement may be used to establish such
an electric field within drift tube 40 in the direction
of arrow 55. It is to be understood, however, that a
constant electric field in the direction of arrow 55 is

20 established to accelerate positively charged ions toward
tube end 44, and that such an electric field may be
reversed to thereby accelerate negatively charged ions
toward tube end 44.

Drift tube 40 may optionally be surrounded by a

25 variable temperature housing 58 which is connected to a
variable temperature source 60 via path 62, all of which
are shown in phantom. In one embodiment, variable
temperature source 60 is a fluid holding tank and path
62 is a conduit leading to housing 58 which, in this

30 case, is preferably sealed. A return conduit (not
shown) is also connected to the fluid holding tank so

that fluid from within the tank may be circulated
through housing 58. The fluid within the fluid holding
tank may be a heated or cooled gas or liquid such as,
for example, liquid nitrogen. In an alternate

5   embodiment, variable temperature source 60 is a known
electrically actuatable temperature controller, and path
62 comprises a pair of electrical conductors connected
between the controller and housing 58. In operation,
temperature controller 60 is operable to heat or cool

10   housing 58 as desired. Regardless of the particular
embodiment of housing 58, source 60 and path 62, the
present invention contemplates that source 60 may
furthermore be controlled by computer 38 via signal path
64.

15   Drift tube 40 is further surrounded by a housing 70
which defines a tube end 66 covering an ion entrance end
thereof, wherein tube end 66 defines an opening, or ion
aperture, 68 therethrough, and an ion exit opening, or
aperture, 84 adjacent to endplate 43. Preferably, ion

20   optics 47 are positioned between openings 45 and 84 to
focus ions exiting opening 45 into an ion acceleration
region of TOFMS 36. Openings 45, 68 and 84 are
preferably bisected by drift tube axis 72. An ion
source 74, which will be described more fully

25   hereinafter, is positioned within ion source region 32
and is operable, preferably under the control of
computer 38 via a number, N, of signal paths 76, wherein
N may be any positive integer, to direct ions within the
spectrometer 34 via opening 68. Ions entering drift

30   tube 40 separate in time as a function of their
individual mobilities, as discussed hereinabove, and are

sequentially directed through opening 70 toward TOFMS
36.

Housing 70 includes a pump 80 for controlling the
pressure of the buffer gas. Preferably, pump 80 is a
5   diffusion pump, the operation of which may be controlled
by computer 38 via signal path 82. Alternatively, pump
80 may be manually controlled by a manual pump actuator
(not shown). In any case, pump 80 is operable to
establish a desired pressure of the static buffer gas
10   within drift tube 40. In accordance with known IMS
techniques, the buffer gas within drift tube 40 may
typically be set within the range of between
approximately one and a few thousand Torr.

TOFMS 36 is preferably surrounded by a housing 126
15   that is attached to IMS 34. TOFMS 36 includes a first
electrically conductive grid or plate 86 connected to a
second voltage source VS2 88 via signal path 90, which
is preferably controlled by computer 38 via signal path
92. A second electrically conductive grid or plate 94
20   is connected to a third voltage source VS3 96 via signal
path 98, which is preferably controlled by computer 38
via signal path 100. A third electrically conductive
grid or plate 102 is connected to a fourth voltage
source VS4 via signal path 106, which is preferably
25   controlled by computer 38 via signal path 108. Grids or
plates 86, 94 and 102 define first and second ion
acceleration regions therebetween as is known in the
art, and which will be more fully described hereinafter.
Those skilled in the art will recognize that other known
30   ion acceleration region structures may be used with

TOFMS 36, such as, for example, positioning a fourth grid or plate between grids or plates 94 and 102.

Grid or plate 102 has a plate surface attached to one end of a flight tube 110, the opposite end of which

5   is attached to a surface of a fourth electrically conductive grid or plate 112. An ion detector 116 is disposed adjacent to grid or plate 112 with an air gap 114 defined therebetween. Ion detector 116 is connected to a fifth voltage source VS5 118 via signal path 120,

10 which is preferably controlled by computer 38 via signal path 122. Ion detector 116 further has a signal output connected to computer 38 via signal path 124, whereby detector 116 is operable to provide ion arrival time information to computer 38. Grids or plates 86, 94, 102

15 and 112 are preferably arranged in juxtaposition with each other such that all plate surfaces having greatest surface area are parallel with each other as well as to the surface of the ion detector 116, and are further preferably perpendicular to a longitudinal axis 128

20 defined centrally through the flight tube 110, which will hereinafter be referred to as the flight tube axis 128.

TOFMS 36 further includes a pump 130 for controlling the vacuum of the TOFMS chamber defined by

25 housing 126. Preferably, pump 130 is a diffusion pump, the operation of which may be controlled by computer 38 via signal path 132. Alternatively, pump 130 may be manually controlled by a manual pump actuator (not shown). In any case, pump 130 is operable to establish

30 a desired vacuum within housing 126 which may be set, in accordance with know TOFMS operating techniques, to

20

within the range of between approximately 10^{-4} and 10^{-10}
Torr.

In the instrument 30 illustrated in FIG. 4, TOFMS
36 is preferably arranged relative to IMS 34 such that
5   the flight tube axis 128 is perpendicular to the drift
tube axis 72. Moreover, TOFMS 36 is preferably
positioned relative to IMS 34 such that the drift tube
axis 72 and the flight tube axis 128 bisect within the
first ion acceleration region defined between grids or
10   plates j86 and 94. In an alternative configuration of
TOFMS 36, grid or plate 94 may be omitted, and the TOFMS
36 need then be positioned relative to IMS 34 such that
the drift tube axis 72 bisects the flight tube axis 128
within the ion acceleration region defined between grids
15   or plates 86 and 102. In either case, TOFMS is
preferably positioned relative to IMS 34 such that the
drift tube axis 72 bisects the flight tube axis 128
approximately centrally within the region of interest.

In the operation of instrument 30, ions are
20   generated by ion source 74, in accordance with one or
more ion generation techniques described hereinafter,
and are supplied to IMS 34 via IMS inlet opening 68. A
buffer gas typically used in IMS instruments 34 is
supplied to drift tube 40 via buffer gas source 46,
25   wherein the buffer gas is regulated to a desired
pressure via pump 80, buffer gas source 46 or a
combination thereof. Typically, the buffer gas is
regulated to a pressure of between approximately 1 and a
few thousand Torr. Voltage source 52 supplies a voltage
30   sufficient to generate a constant electric field along

the drift tube axis in a direction indicated by arrow
55.

In accordance with known IMS 34 operation, ions
entering IMS inlet opening 68 travel through drift tube

5   40 toward IMS outlet opening 84, wherein the ions
separate in time according to their individual
mobilities. Ions having low mobility lag behind those
having higher mobility, wherein ion mobilities are
largely a function of their collision cross-sections.

10   As a result, the more compact ions arrive at the IMS
outlet opening 84 more quickly than more diffuse ions.
Those skilled in the art will recognize that the
temperature of drift tube 40 may also be controlled via
variable temperature source 60 so that ion mobility

15   analysis may be performed as a function of temperature.

TOFMS 36 is operable to accelerate ions from the
space defined between grids or plates 86 and 94 toward a
field-free flight tube 110, wherein the ions separate in
time according to their individual masses. Generally,

20   ions having less mass will reach the detector 116 more
quickly than those having greater mass. The detector
116 is operable to detect arrival times of the ions
threat and provide signals corresponding thereto to
computer 38 via signal path 124.

25   As set forth in greater detail in U.S. Patent Nos.
5,504,326, 5,510,613 and 5,712,479 to Reilly et al.,
which have been incorporated herein by reference,
voltage sources VS2 88, VS3 96 and VS4 104 are typically
controlled by computer 38 to initially establish

30   voltages at grids or plates 86, 94 and 102 that match
the voltage level associated with IMS 34 (which is set

by voltage source VS1 52). Depending upon various
instrument parameters, such as the length of flight tube
110, the distances between grids or plates 88, 94, 102
and 112, and the distance 114 between grid or plate 112

5   and detector 116, as well as estimates of initial ion
position or initial ion velocity within the space
defined between grids or plates 86 and 94, computer 38
is operable to control sources 88, 96 and/or 104 to
instantaneously increase the electric field between

10 grids or plates 86, 94 and 102 to thereby create an ion
drawout electric field therebetween which accelerates
ions between these grids toward flight tube 110.
Preferably, the pulsed ion drawout electric field is in
a direction from grid or plate 86 toward flight tube 110

15 to thereby accelerate positively charged ions toward the
flight tube 110. Those skilled in the art will
recognize, however, that this electric field may
alternatively be reversed to accelerate negatively
charged ions toward the flight tube 110.

20   In any event, ions within the space defined between
grids or plates 86 and 94 are accelerated by the pulsed
ion drawout electric field to the space defined between
grids or plates 94 and 102. Due to the fact that ions
entering the region defined between grids or plates 86

25 and 94 along axis 72 have a narrow spatial distribution,
due to focusing of the ions into this region via ion
optics 47, and a small velocity component along axis
128, it is possible to choose the pulsed voltage applied
to grids or plates 86 and/or 94 in such a way as to

30 obtain sharp TOFMS peaks. The goal of the pulsed ion
drawout electric field and the subsequent acceleration

of the ions between grids or plates 94 and 102 is to
provide all ions reaching grid or plate 102 with
substantially the same kinetic energy. The flight tube
110 has no electric field associated therewith so that
5   the ions drift from grid or plate 102 toward detector
116, wherein the ions separate in time as a function of
their individual masses as described hereinabove.
Computer 38 typically controls voltage source VS5 118
to supply a voltage thereto during detection times to
10   thereby increase the gain of detector 116 as is known in
the art. Pump 130 controls the vacuum within TOFMS 36,
and pump 130 is preferably controlled by computer 38 via
signal path 132. TOFMS 36 is typically operated between
10^{-4} and 10^{-10} Torr.

15   In the embodiment 30 of the hybrid IMS/TOFMS
instrument illustrated in FIG. 4, drift tube axis 72
preferably bisects the space defined between grids or
plates 86 and 94 of TOFMS 36, and is perpendicular to
flight tube axis 128. The present invention
20   alternatively contemplates arranging TOFMS 36 relative
to IMS 34 such that the drift tube axis 72 passes
between grids or plates 86 and 94 perpendicular to
flight tube axis 128, but at some other known distance
relative to either of the grids or plates 86 and 94. In
25   either case, the foregoing structural positioning of
TOFMS 36 relative to IMS 34 provides advantages over
non-perpendicular arrangements of the drift tube axis 72
relative to the flight tube axis 128. For example, such
a perpendicular arrangement ensures that ion packets
30   entering the ion acceleration region defined between
grids or plates 86 and 94 from IMS 34 will have constant

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and relatively well defined initial ion positions as
they travel therebetween along axis 72. As discussed
briefly hereinabove, ion optics 47 focus ions into the
ion acceleration region to thereby minimize spatial
5   distribution of the ions. Moreover, since axis 72 is
parallel with grids or plates 86 and o4, ion position
with respect to axis 128 will remain relatively
constant. This feature provides for the ability to
accurately estimate initial ion position within the ion
10   acceleration region defined between grids or plates 86
and 94, to thereby allow a more accurate estimation of
the pulsed ion drawout electric field discussed above.

       Preferably, computer 38 controls the generation of
ions from ion source 74, as will be discussed in greater
15   detail hereinafter, so that computer 38 has knowledge of
the times at which ions were introduced into IMS 34,
hereinafter referred to as ion introduction events. The
computer 38 is then operable to control voltage sources
88 and 96 to repeatedly provide the pulsed ion drawout
20   field some number of times for every ion introduction
event. In one embodiment, a pulsed ion drawout field is
repeatedly provided 512 times for every ion introduction
event. Those skilled in the art will recognize that the
number of pulsed ion drawout fields provided for every
25   ion introduction event is directly proportional to the
ultimate resolution of the instrument 30. As this
pulsed operation relates to some of the advantages of
the perpendicular positioning of TOFMS 36 relative to
IMS 34, such an arrangement minimizes the possibility
30   that all or part of any one ion packet will travel
through the TOFMS 36 unprocessed. Due to the direction

of travel of the ion packets relative to the grids or
plates 86 and 94, and also to the pulsed nature of the
ion drawout electric field, the TOFMS 36 will have
multiple chances to accelerate each ion packet toward
5   detector 116 as they travel along axis 72. As such, the
instrument 30 is configured to provide for maximum ion
throughput to detector 116.

Referring now to FIG. 5, an alternate embodiment of
a hybrid ion mobility and time-of-flight mass
10   spectrometer 150, in accordance with the present
invention, is shown. Spectrometer 150 is similar in
many respects to spectrometer 30 shown in FIG. 4 and
described hereinabove, and like components are therefore
identified with like numbers. Discussion of the common
15   components, as well as the basic operation of IMS 34 and
TOFMS 36', will therefore not be repeated for brevity's
sake.

Unlike instrument 30 of FIG. 4, the TOFMS 36' of
instrument 150 is positioned relative to IMS 34 such
20   that the drift tube axis 72 also defines the flight tube
axis of TOFMS 36'. Alternatively, TOFMS 36' could be
arranged relative to IMS 34 with any orientation such
that the drift tube axis 72 is non-perpendicular to the
flight tube axis. In any such orientation, the initial
25   positions of the ion packets within the space defined
between grids or plates 86' and 94 either cannot be
estimated with any degree of accuracy (as in the
orientation illustrated) or changes as the ion packets
travel along axis 72 (as in any non-perpendicular
30   arrangement). Moreover, in any such orientation, it is
difficult to estimate when, relative to an ion

introduction event, the ion packets will arrive within

the space defined between grids or plates 86' and 94,

and the timing of the pulsed ion drawout electric fields

is thus difficult to predict. As a result, it is likely

5   that the timing of the pulsed ion drawout electric

fields will be inaccurate so that ions may be lost

within the TOFMS 36' and/or the mass resolution of the

TOFMS 36' will be adversely affected.

In order to address the foregoing problems

10   associated with non-perpendicular positioning of the

TOFMS 36' relative to the IMS 34, which are the same

problems associated with the Guevremont et al. system

discussed hereinabove in the BACKGROUND section,

instrument 150 is provided with an ion trap 152

15   operatively positioned between the ion outlet opening 84

of IMS 34 and the space defined between grids or plates

86' and 94. In the embodiment illustrated in FIG. 5,

grid or plate 86' defines an ion inlet opening 178

therethrough which is aligned along axis 72 with ion

20   outlet opening 84 of IMS 34. In other non-perpendicular

arrangements of TOFMS 36' relative to IMS 34, ion inlet

opening 178 may not be required since ions may enter the

space between grids or plates j86' and 94 in the same

manner as discussed with respect to the embodiment 30

25   illustrated in FIG. 4.

In any event, ion trap 152 is preferably a known

quadrupole ion trap having a first endcap 154, a center

ring 162 and a second endcap 170. Each of the endcaps

154 and 170 define apertures therethrough which align

30   with axis 72. In this configuration, ion trap 152

confines ions therein to a small volume in its center

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which is in alignment with the ion inlet opening to
TOFMS 36'. First endcap 154 is connected to a voltage
source VS6 156 via signal path 158, which is itself
connected to computer 38 via signal path 160. Center

5   ring 162 is connected to a voltage source VS7 164 via
signal path 166, which is itself connected to computer
38 via signal path 168, and second endcap 170 is
connected to a voltage source VS8 172 via signal path
174, wherein source 172 is connected to computer 38 via

10 signal path 176. Preferably, sources 156 and 172 are
operable to produce DC voltages and source 164 is
operable to produce AC voltages in the RF range.

In operation, computer 38 controls sources 156 and
172 to bias endcaps 154 and 170 such that ions exiting

15 ion outlet opening 84 of IMS 34 have just enough energy
to enter the opening defined in the first endcap 154.
Once therein, the ions collide with buffer gas leaking
out of opening 84 into the trap 152, and lose sufficient
energy thereby so that the RF voltage on center ring 162

20 is operable to confine the ions within the trap 152.
The confined ions undergo further collisions inside the
trap 152 which causes the ions to correspondingly
experience further energy loss, resulting in a
concentration of the ions toward the center of ring 162

25 due to the RF voltage thereon. As long as the voltages
on endcaps 154 and 170 and center ring 162 are
maintained, ions may enter the trap 152 and collect
therein. Ions are ejected out of the trap 152 by
turning off the RF voltage on center ring 162 and

30 applying an appropriate DC pulse to one of the endcaps
154 or 170. For example, to eject a collection of

positively charged ions from trap 152, either the
voltage on endcap 154 may be pulsed above that present
on endcap 170 or the voltage on endcap 170 may be pulsed
below that present on endcap 154. In general, the

5   magnitude of the RF field applied to the center ring via
source 164, as well as any DC voltage included therein,
may be varied to thereby select ions of any desired mass
to charge ratio to be collected by ion trap 152. Ions
of all mass to charge ratios, or ions of any particular

10   mass to charge ratio, may be selectively collected
within ion trap 152 through proper choice of DC level
and RF peak magnitude provided by voltage source 164.

     As it relates to the present invention, the ion
trap 152 is controllable by computer 38 to periodically

15   eject the collected ion packets therefrom, hereinafter
referred to as an ion ejection event, so as to provide
for a more accurate estimate of initial ion position
within the space defined between grids or plates 86'and
94. Since the computer 38 controls the time at which a

20   packet of collected ions is ejected from ion trap 152,
the time at which the ion packet arrives at a specified
position in the space defined between grids or plates
86' and 94 can be accurately estimated. Knowing the
approximate time, relative to the ion ejection event, at

25   which the ion packet arrives at the specified position
between grids or plates 86' and 94, computer 38 may more
accurately estimate appropriate timing for applications
of the pulsed ion drawout electric field to thereby
provide for maximum mass resolution as discussed

30   hereinabove. Moreover, providing for a more accurate
estimate of the timing of the pulsed ion drawout

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electric fields reduces the likelihood that ion packets, or at least portions thereof, will be lost within the TOFMS 36'.

In the operation of instrument 150, IMS 34 is
5   operable to provide packets of ions, which are separated in time as a function of ion mobility, to TOFMS 36' via ion outlet opening 84. Computer 38 controls ion trap 152 to collect the various ion packets therein one at a time, and eject each collected ion packet therefrom at
10 periodic intervals. The ejected ions enter the space defined between grids or plates 86' and 94 as discussed hereinabove, and computer 38 is operable to computer appropriate times at which to apply the pulsed ion drawout electric fields based on the timing of the ion
15 ejection events. The TOFMS 36' is thereafter operable as described hereinabove to produce mass spectrum information.

Referring now to FIG. 6, a plot 190 of ion flight time vs. ion drift time for an oligothymidine sample is
20 shown, wherein the data shown is producible via either instrument embodiment 30 or 150. As compared to the plot of FIG. 3, it is apparent that the hybrid ion mobility and time-of-flight mass spectrometer of the present invention is operable to resolve structural
25 information of molecules in two substantially orthogonal dimensions. For each drift time, corresponding to arrival in the TOFMS of a corresponding ion packet, the instrument of the present invention is operable to resolve a number of times-of-flight, corresponding to a
30 number of mass to charge ratios. The plot 190 of FIG. 6 thus illustrates that the total resolving power of

instrument 30 is drastically better than that achievable

via an IMS or TOFMS alone. This technique dramatically

reduces the problem of congestion of mass spectra, due

to mass peak overlap, in obtaining sequence information

5   for large biomolecules (in excess of 50 residues). The

present invention thus provides an instrument for

composition, sequence and structural analysis of

biomolecules which does not suffer from drawbacks

associated with prior art systems discussed in the

10   BACKGROUND section.

Referring now to FIG. 7A, one preferred embodiment

74' of an ion source 74 for either of the instrument

embodiments of FIGS. 4 and 5, is shown. Embodiment 74'

includes a chamber 200 having a sample 202 mounted

15   therein and an optical window 206 extending therefrom.

A radiation source 204 is electrically connected to

computer 38 via signal path 76A, and is configured to

direct radiation through optical window 206 to thereby

irradiate sample 202. Chamber 200 may include a conduit

20   extending therefrom to a pump 208 which may be

controlled by computer 38 via signal path 76B.

Ion source 74' is a known MALDI arrangement wherein

radiation source 204, preferably a laser, is operable to

desorb gaseous ions from a surface of the sample 202.

25   Computer 38 is operable to control activation times of

laser 204 to thereby control sample ionization events.

The desorbed ions are directed by the internal structure

of chamber 202 to ion inlet opening 68 of IMS 34. The

sample 202 may, in accordance with the present

30   invention, be a biomolecule of any size such as DNA,

RNA, any of various proteins, carbohydrates,

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glycoconjugates, and the like. Pump 208 may be controlled to pressurize chamber 208 to thereby conduct high pressure MALDI analysis as is known in the art.

5   Referring now to FIG. 7B, an alternate embodiment 74'' of an ion source 74 for either of the instrument embodiments of FIGS. 4 and 5, is shown. Embodiment 74'' includes a liquefied sample 220 having a spray hose or nozzle 222 extending toward an opening defined in a desolvation region 226. Actuation of the spray nozzle 10   222 may be manually controlled, as is known in the art, or may be controlled by computer 38 via signal path 76C. Desolvation region 226 is connected to computer 38 via signal path 76C', and is operable to convert charged sample droplets supplied thereto via nozzle 222 into 15   gaseous ions and supply these ions to a ion optics member 228. Optics member 230 is operable to focus the gaseous ions and direct them into ion inlet opening of IMS 34. Ion source region 32 includes a conduit extending therefrom to a pump 232 which may be 20   controlled by computer 38 via signal path 76D.

Ion source 74'' is a known electrospray ionization (ESI) arrangement operable to convert a liquefied solution containing the sample to gaseous ions. Computer 38 is operable to control activation times of 25   desolvation region 226 to thereby control sample ionization events. Pump 232 is operable to pressurize the ion source region 32 as is known in the art, and the desolvation region 226 is operable convert the liquefied solution to gaseous ions. The sample source 220 may, in 30   accordance with the present invention, include a solution containing a biomolecule of any size such as

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DNA, RNA, any of various proteins, carbohydrates, glycoconjugates, and the like.

Referring now to FIG. 7C, another alternate embodiment 74′′′ of an ion source 74 for either of the instrument embodiments of FIGS. 4 and 5, is shown. Embodiment 74′′′ includes a sample source 236, which may be either of the foregoing sample sources 74′ or 74′′ illustrated in FIGS. 7A or 7B, and which may be controlled as described hereinabove by computer 38 via a number, M, of signal paths 76E, wherein M may be any integer less than N (see FIGS. 4 and 5).

Ion source 74′′′ further includes an ion trap 152 positioned between ion source 236 and the ion inlet opening 68 of IMS 34. Ion trap 152 is preferably a known quadrupole ion trap identical to that shown in FIG. 5 and described hereinabove. A detailed discussion of the operation of ion trap 152 therefore need not be repeated here. Endcap 154 is connected to a voltage source VS9 238 via signal path 240, center ring 162 is connected to a voltage source VS10 242 via signal path 244 and endcap 170 is connected to a voltage source VS11 246 via signal path 248. VS9, VS10 and VS11 are each connected to computer 38 via signal paths 76F, 76G and 76H, respectively. Computer 38 is operable to control VS9, VS10 and VS11 identically as described with respect to VS6, VS7 and VS8, respectively, of FIG. 5.

In operation, computer 38 is operable to control ion trap 152, in a manner similar to that described hereinabove, to collect a bulk of ions therein and selectively eject the collected ions therefrom toward ion inlet opening 68 of IMS 34. As is known in the art,

the peak resolution of an ion mobility instrument, such
IMS 34, is limited by the length of the input pulse of
ions into the instrument. Generally, mobility peaks
cannot be resolved any better than the time length of

5   the input ion pulse. A drawback particularly associated
with the use of ESI is that the input ion pulse width
must typically be at least 50 µs in order to produce
enough ions for analysis. However, with the ion source
arrangement 74′′′ shown in FIG. 7C, computer 38 is

10 operable to collect a large number of ions within ion
trap 152 prior to pulsing the ions into the IMS 34.
With a sufficient number of ions collected in ion trap
34, the only limitation on the ion input pulse length,
and hence the resolution capability of IMS 34, is the

15 time required to open and close ion trap 152. With
existing ion traps, the ion input pulse lengths may be
reduced to less than 1.0 µs in duration.

FIGS. 8A and 8B show a comparison of ion mobility
distributions for a maltotetraose sample, wherein the

20 spectrum 250 of FIG. 8A was produced using an ESI source
similar to that shown in FIG. 7B, with 100,083 input
pulses of 20 µs duration. The spectrum 252 of FIG. 8B
was produced using the same ESI source as that used for
FIG. 8A along with an ion trap, such as ion trap 152

25 shown in FIG. 7C, with 4003 pulses of 1 µs duration.
Compared to spectrum 250, spectrum 252 has a 4-5 times
increase in signal strength, an increase in resolution
by a factor of approximately 20 and an increase in
signal-to-noise ratio by a factor of approximately 20 as

30 well.

Referring again to FIG. 7C, ion trap 152 may be
used with any known ion generation source to increase
not only the resolution and sensitivity of IMS 34 along,
but also the resolution and sensitivity of either hybrid
5   instrument 30 or 150 of FIGS. 4 and 5.

It is to be understood that either embodiment of
the hybrid ion mobility and time-of-flight mass
spectrometer shown and described herein is capable of
operation in a number of different operational modes.
10   For example, the structure and operation of the various
embodiments of the present invention have been described
herein according to a first mode of operation wherein
ions of relatively low energy are generated and injected
into the hybrid instrument, from which structural
15   information relating to the ions can be obtained.

In a second mode of operation, such ions could be
injected into the hybrid instrument at higher energies,
wherein high energy collisions with the buffer gas
within the IMS 34 result in ion fragmentation. In such
20   a case, the ion fragments, separated in time as a
function of their mobilities, would be supplied to the
TOFMS portion of the instrument, wherein mass spectra
information of the various fragments could be obtained
for sequencing analysis. Alternatively, fragmentation
25   of ions for such analysis may be accomplished via any of
a number of other known techniques. Examples of such
known alternative ion fragmentation techniques include
enzyme degradation fragmentation, photo-fragmentation,
thermal dissociation such as by heating drift tube 40
30   via control of variable temperature source 60, electron

impact dissociation, surface induced dissociation, and blackbody infrared radiation induced dissociation.

In a third mode of operation, ions of only a particular mass could be processed by the hybrid

5   instrument. One way of generating ions of only a particular mass is to adjust the peak amplitude and/or DC voltage of the center ring voltage source of an ion trap positioned prior to the IMS 34. By properly adjusting this voltage, ion trap 152 may be configured

10   to store therein only ions having a particular mass to charge ratio. In this manner, the ion trap 152 is controlled to act as an ion filter. Another way of analyzing ions of only a particular mass is to provide an ion trap 152 between the IMS 34 and TOFMS 36, and

15   controlling the ion trap 152 as just discussed to filter out ions having undesirable mass to charge ratios.

In a fourth mode of operation, high energy ions of only a particular mass are introduced into the IMS 34. Therein, these ions undergo fragmentation, and such

20   fragments could then be further processed by the TOFMS 36 as discussed above.

Referring now to FIG. 9, one preferred embodiment of an ion mobility and mass spectrometer instrument 300 that is particularly well suited for conducting

25   sequencing analysis in a manner similar to that just described hereinabove with respect to the second mode of operation, in accordance with the present invention, is shown. Several of the components of instrument 300 are identical to those shown and described with respect to

30   FIGS. 4 and 5, and some of the structural and operational details thereof will accordingly be omitted

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here for brevity. For example, instrument 300 includes
an ion source 32 operatively connected to an ion
mobility spectrometer (IMS), wherein IMS 34 includes a
source of buffer gas 46 that is controllable via
5   operation of a pump 80 as described hereinabove.
Instrument 300 further includes a mass spectrometer (MS)
36, preferably a time-of-flight mass spectrometer
(TOFMS), that is configured to receive ions from IMS 34
as described hereinabove. In this embodiment, however,
10   the drift tube axis of IMS 34 (not shown in FIG. 9) and
the flight tube axis of TOFMS 36 (not shown in FIG. 9)
may be arranged at any desired angle with respect to
each other. It has been determined through
experimentation that for non-perpendicular
15   configurations of IMS 34 relative to TOFMS 36 (i.e.,
configurations other than that illustrated in FIG. 4),
an ion trap 152 (see FIG. 5) is not required as
described hereinabove if the ion acceleration region
(between grids 86, 94 and 102) of TOFMS 36 is
20   continually activated or pulsed. In other words, ions
need not be collected in an ion trap 152 for timing
purposes if the ion acceleration region of TOFMS 36 is
continually pulsed in a free-running operational mode.
Accordingly, ion trap 152 may be omitted from any
25   perpendicular or non-perpendicular configurations of the
IMS drift tube axis relative to the TOFMS flight tube
axis, although the present invention contemplates that
such an ion trap 152 may optionally be used in such
configurations as desired, wherein trap 152 may be
30   positioned adjacent to the entrance of TOFMS 36.

Instrument 300 further includes a computer 310 having a memory 312. Computer 310 is preferably operable to control the flow rate of buffer gas #1 within buffer gas source 46 via signal path 48, and is

5   further preferably operable to control pump 80 of IMS 34 via signal path 82 and a vacuum pump 130 of TOFMS 36 via signal path 132, as described hereinabove. Computer 310 is also operable to control ion source 32 via a number, N, of signal paths 76, wherein N may be any integer, and

10 is further operable to receive ion detection signals from TOFMS 36 via signal path 124 and process such signals to produce two-dimensional ion spectra; e.g. ion mass vs. ion mobility, as described hereinabove.

Instrument 300 includes a number, J, of voltage

15 sources 314_1 - 314_J connected to computer 310 via signal paths 316_1 - 316_J. Voltage sources 314_1 - 314_J are operatively connected to IMS 34 via corresponding signal paths 318_1 - 318_J. In operation, computer 310 is operable to control voltage sources 314_1 - 314_J to

20 thereby control the operation of IMS 34 as described hereinabove. Instrument 300 further includes another number, M, of voltage sources 330_1 - 330_M connected to computer 310 via signal paths 332_1 - 332_M. Voltage sources 330_1 - 330_M are operatively connected to TOFMS 36

25 via corresponding signal paths 334_1 - 334_M. In operation, computer 310 is operable to control voltage sources 330_1 - 330_M to thereby control the operation of TOFMS 36 as described hereinabove.

The components of instrument 300 described thus far

30 with respect to FIG. 9 are identical to previously described components of the instruments 30 and/or 150 of

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FIGS. 4 and 5. Unlike instruments 30 and 150, however, instrument 300 further includes a quadrupole mass filter 302 having an ion inlet coupled to the ion outlet of IMS 34 and an ion outlet coupled to an ion inlet of a

5   collision cell 304 of known construction. An ion outlet of collision cell 304 is coupled to an ion inlet of TOFMS 36; i.e., to the ion acceleration region defined between plates or grids 86 and 94 of TOFMS as shown in FIGS. 4 and 5. Collision cell 304 includes a source of

10   buffer gas 306, wherein the flow rate of buffer gas #2 is controlled by computer 310 via signal path 307, preferably in a manner described hereinabove with respect to the computer control of the buffer gas source 46 of FIG. 4. Alternatively, buffer gas source 306 may

15   be omitted and buffer gas source 46 may be configured to provide buffer gas #1 to cell 304 via conduit 305 as shown in phantom in FIG. 9. Collision cell 304 further includes a pump 308 of known construction, the operation of which is controlled by computer 310 via signal path

20   309. As is known in the art, pump 308 may be controlled to establish and maintain a desired quantity of buffer gas within collision cell 304, and may further be controlled to purge cell 304 of buffer gas. Alternatively, structure 308 may represent a manually

25   actuatable or computer controlled valve. In this case, valve 308 may be controlled to establish and maintain a desired quantity of buffer gas #2 within collision cell 304, or may alternatively be controlled to establish and maintain a desired quantity of buffer gas #1 within the

30   quadrupole mass filter 302 and collision cell 304.

A number, K, of voltage sources 320_1 – 320_K are
provided, wherein K may be any integer, and wherein
control inputs of sources 320_1 – 320_K are connected to
computer 310 via corresponding signal paths 322_1 – 322_K.
5   Outputs of voltage sources 320_1 – 320_K are operatively
connected to the quadrupole mass filter (QMF) 302, in a
manner to be described more fully hereinafter with
respect to FIGS. 11 and 12, via corresponding signal
paths 324_1 – 324_K.   A number, L, of voltage sources 326_1
10   – 326_L are provided, wherein L may be any integer, and
wherein control inputs of sources 326_1 – 326_L are
connected to computer 310 via corresponding signal paths
328_1 – 328_L.   Outputs of voltage sources 326_1 – 326_L are
operatively connected to the collision cell 304 in a
15   known manner via corresponding signal paths 329_1 – 329_L.

Referring now to FIG. 10, a cross-section of
another preferred structure of the ion source 32 for use
with any of the instruments illustrated in FIGS. 4, 5
and 9, in accordance with the present invention, is
20   shown.   Ion source 32 includes an ion source chamber 350
separated from an ion collection chamber 354 by a wall
or partition 355.   Ion source chamber 350 includes a
port having a conduit 352 connected thereto, wherein
conduit 352 is preferably connected to a pump or valve
25   of known construction for changing gas pressure within
region 350.   An ion source 74 is disposed within region
350, wherein source 74 may be any of the ion sources
74', 74" or 74''' described hereinabove with respect to
FIGS. 7A-7C, and/or any combination thereof.   Wall or
30   partition 355 includes an aperture 353 therethrough that
is aligned with an ion outlet of ion source 74 and is

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also preferably aligned with a longitudinal axis of the
drift tube 40 of IMS 34, wherein aperture 353 defines an
ion inlet to ion collection chamber 354. An
electrically conductive grid, or series of vertically or
5   horizontally parallel wires, 356 (hereinafter "grid") is
positioned across the ion inlet aperture 68 of IMS 34,
wherein grid 356 is connected to one of the voltage
sources 314_1 via signal path 318_1. Computer 310 is
operable to control the voltage of grid 356, as is known
10   in the art, to thereby permit and inhibit entrance of
ions into IMS 34. For example, computer 310 is operable
to inhibit entrance of ions into IMS 34 by activating
voltage source 314_1 to thereby cause ions in the vicinity
of grid 356 to be attracted thereto and neutralized upon
15   contact. Conversely, computer 310 is operable to permit
entrance of ions into IMS 34 by deactivating voltage
source 314_1 to thereby permit passage of ions
therethrough. Alternatively, the ion gating function
may be accomplished by a voltage source 320_2 connected to
20   guard rings 50 via signal path 318_2, wherein computer 310
is operable to control source 320_2 to attract ions to
guard rings 50 when it is desirable to inhibit ions from
traveling through drift tube 40. In this case, grid 356
and voltage source 320_1 may be omitted from FIG. 10.
25   Alternatively still, the ion gating function may be
accomplished by impressing a voltage across aperture 68
to thereby create an electric field therebetween. In
this case, computer 310 is operable to control the
voltage across aperture 68 to divert ions toward guard
30   rings 50 when it is desirable to inhibit ions from
traveling through drift tube 40. Those skilled in the

art will recognize that any known technique for pulsing ions from ion collection chamber 354 through ion inlet aperture 68, including for example any known electrical, mechanical and/or electro-mechanical means, may be used,

5   and that any such technique falls within the scope of the present invention.

In any case, the ion collection chamber 354 is functionally similar to the ion trap 152 of FIG. 7C in that it provides for the collection of a large quantity

10 of ions generated by ion source 74 prior to entrance into IMS 34. Through appropriate control of ion source 74 and grid 356 or equivalent, the quantity of ions entering IMS 34 may thus be correspondingly controlled.

Referring now to FIG. 11, a cross-section of the

15 quadrupole mass filter (QMF) 302, as viewed along section lines 11-11 of FIG. 9, is shown. QMF 302 includes four electrically conductive rods or plates 360, 362, 364 and 366 that are preferably disposed equidistant from a longitudinal axis 365 extending

20 through QMF 302. Two of the opposing rods 360 and 362 are electrically connected to voltage source 320_1 via signal path 324_1, wherein source 320_1 has a control input connected to computer 310 via signal path 322_1. Signal path 324_1 is connected to a signal phase shifter 366 of

25 known construction via signal path 368, wherein a signal output of phase shifter 366 is electrically connected to the remaining two opposing rods 364 and 366. Computer 310 is operable to control voltage supply 320_1, which is preferably a radio frequency (RF) voltage source, to

30 thereby control the RF voltage applied to rods 360 and 362. Phase shifter 366 is preferably operable to shift

the phase of the RF voltage on signal path 368 by 180°
and apply this phase shifted RF voltage to signal path
324_2. Those skilled in the art will recognize that phase
shifter 366 may alternatively be replaced with a second

5   RF voltage source that is controllable by computer 310
to produce an RF voltage identical to that produced by
source 320_1 except shifted in phase by 180°. In any
case, signal paths 324_1 and 324_2 are electrically
connected to voltage source 320_2 via signal paths 324_3

10 and 324_4 respectively, wherein source 320_2 has a control
input connected to computer 310 via signal path 322_2.
Voltage source 320_2 is preferably a DC voltage supply
controllable by computer 310 to thereby impress a DC
voltage between rod pairs 360/362 and 364/366.

15   In the operation of QMF 302, the RF voltages
applied to rods 360-366 alternately attract ions to rod
pairs 360/362 and 364/366, wherein this attraction
increases with decreasing ion mass-to-charge ratio
(m/z). Below some threshold m/z value (i.e., lighter

20 ions), the ions come into contact with one of the rods
360-366 and are accordingly neutralized or ejected. The
m/z value below which ions are neutralized is determined
by the strength and frequency of the RF signal as is
known in the art. The DC voltage applied to rods 360-

25 366 similarly attracts ions thereto wherein this
attraction increases with increasing m/z values. Above
some threshold m/z value (i.e., heavier ions), the ions
come into contact with one of the rods 360-366 and are
accordingly neutralized. The m/z value above which ions

30 are neutralized is determined by the strength of the DC

signal as is known in the art. Referring to FIG. 12, a
plot 370 of ion intensity at the ion outlet of QMF 302
is shown demonstrating that the RF and DC voltages
applied to rods 360-366 result in passage through QMF

5   302 only of ions having m/z values above a minimum m/z
value m/z_1 and below a maximum m/z value m/z_2. QMF 302
thus acts as a bandpass filter wherein the pass band of
m/z values is controlled via computer 310 by controlling
the operating strength and frequency of the RF voltage

10   supply 320_1 and by controlling the operating strength of
the DC voltage supply 320_2. In accordance with an
important aspect of the present invention, computer 310
is operable, under certain operating conditions, to
control the m/z values of ions being passed from IMS 34

15   to the collision cell 304 as will be descried in greater
detail hereinafter.

        The collision cell 304 is of known construction,
and the filling and purging of buffer gas
therein/therefrom is preferably controlled by computer

20   310 in a known manner. Alternatively, the filling and
purging of cell 304 may be manually controlled via known
means. In either case, when cell 304 is filled with
buffer gas, ions provided thereto by QMF 302 undergo
collisions with the buffer gas and fragmentation of

25   parent ions into a number of daughter ions results as is
known in the art. In a preferred embodiment, the
internal structure of the collision cell 304 is similar
to that of the quadrupole mass filter illustrated in
FIG. 11 except that collision cell 304 includes eight

30   rods (poles) rather than four, and is accordingly
referred to as an octopole collision cell. At least one

of the voltage sources 326_1 – 326_L is preferably a RF
voltage source connected between two pairs of four
opposing poles, wherein computer 310 is operable to
control the RF voltage source to thereby concentrate
5   ions centrally therein and provide a low-loss channel or
pipe between QMF 302 and MS 36. The buffer gas for cell
304 may be, for example, Argon, Helium or Xenon,
although the present invention contemplates using other
gases provided to cell 304 via source 306 or 46 as
10 described hereinabove. The present invention
contemplates that collision cell 304 may alternatively
be configured in accordance with any desired trapping
multiple (e.g., quadrupole, hexapole, etc.).
Alternatively still, collision cell 304 may me
15 configured as a non-trapping gas collision cell. In any
event, those skilled in the art will recognize that the
importance of any such collision cell arrangement lies
in its ability to provide for fragmentation of entering
parent ions into daughter ions.
20   Referring now to FIG. 13, one preferred
embodiment of a process 400 for conducting sequencing
analysis using the instrument 300 illustrated in FIG. 9,
in accordance with the present invention, is shown.
Process 400 begins at step 402 where a counter variable
25 A is set equal to an arbitrary initial number (e.g., 1).
Thereafter at step 404, collision cell 304 is purged of
buffer gas either manually or under the control of
computer 310 in a known manner. It is to be understood,
however, that if no buffer gas initially exists in cell
30 304, step 404 may be avoided. Thereafter at step 406,
computer 310 is operable to control QMF 302 so as to

pass ions having any m/z value therethrough.   In one
embodiment, computer 310 is operable to execute step 406
by deactivating voltage sources 320_1 and 320_2 to thereby
operate QMF 302 in an all-pass operational mode; i.e.,

5   such that QMF 302 passes ions having all m/z values
therethrough.

        Process 400 continues from step 406 at step 408
where computer 310 is operable to activate ion source 74
to thereby begin the generation of ions from a suitable

10   sample source.   Thereafter at step 410, control computer
310 is operable to pulse ion gate 356 (FIG. 10) for a
predetermined duration to thereby permit entrance of a
gaseous bulk of ions from collection chamber 354 into
IMS 34, and to continually pulse the ion acceleration

15   region of MS 36, as described hereinabove, to thereby
operate MS 36 in a free running mode.   Those skilled in
the art will recognize that when using embodiments of
ion source 32 other than that shown in FIG. 10 (e.g.,
those of FIGS. 7A and 7B), steps 408 and 410 may be

20   combined such that computer 310 is operable to activate
the ion source and supply a gaseous bulk of ions to IMS
34 in a single step.   In any case, process 400 continues
from step 410 at step 412 where a spectrum of ion flight
times (i.e., ion mass) vs. ion drift times (i.e., ion

25   mobilities) resulting from passage of ions through IMS
34 and MS 36, as described hereinabove, is observed.

        Referring now to FIGS. 14A-14D, a graphical example
of steps 410 and 412 is illustrated.   Signal 450 of FIG.
14A represents the voltage at ion gate 356, wherein

30   computer 310 is operable to pulse gate 356 to an
inactive state for a predetermined duration at step 410

to thereby permit entrance of a bulk of gaseous ions
into IMS 34. Signal 452 of FIG. 14B represents the
voltage at the ion acceleration region of TOFMS 36,
wherein computer 310 is operable to pulse the ion

5   acceleration region in a free running manner at step 410
to thereby periodically accelerate ions or parts of ions
toward the ion detector. A typical value for the
duration of deactivation of ion gate signal 450 is 100
μs, a typical value for the duration of activation of

10 the TOFMS signal 452 is 3 μs, and a typical value for
the time between TOFMS signal activation is 100 μs.
However, the present invention contemplates other values
for the foregoing signal durations, and it will be
understood that the actual signal durations used will

15 typically be dictated by many factors including sample
type, analysis mode, information sought and the like.
In any case, signal 454 of FIG. 14C represents the
activation state of QMF 302, wherein computer 310 is
operable throughout steps 410 and 412 to maintain QMF

20 302 in an inactive or all-pass state; i.e. QMF 302 is
operable to pass ions having any m/z value therethrough.
Finally, a spectrum 456 of ion drift time (corresponding
to ion mobility) vs. ion flight time (corresponding to
ion mass) is shown in FIG. 14D illustrating one example

25 of the resulting ion spectrum of step 412.

    Close inspection of spectrum 456 of FIG. 14D
reveals that ions a, b and g do not overlap in drift
times with any other ion, while ions c and d and ions e
and f overlap in their respective drift times. Ions c

30 and d will accordingly arrive at collision cell 304 at

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approximately the same time (3.5 μs), and ions e and f
will accordingly arrive at collision cell 304 at
approximately the same time (4.8 μs). If collision cell
304 was filled with buffer gas so that ion fragmentation

5   occurred, TOFMS 36 would not be able to accurately
distinguish parent and daughter ions attributable to ion
c from those of ion d and likewise those attributable to
ion e from those of ion f. If, however, no such
overlaps occurred, the foregoing problem would not

10   occur. In accordance with an important aspect of the
present invention, process 400 is configured to conduct
subsequent sequencing analysis (via fragmentation) with
QMF 302 operating in an all-pass mode if no overlap in
ion drift times are evident from step 412, but is

15   alternatively operable to conduct subsequent sequencing
analysis (via fragmentation) with QMF 302 operable to
selectively filter out all but one of the ions
overlapping in any one drift time. In the latter case,
the sequencing analysis is repeated until fragmentation

20   spectra are produced for all ions in the original
spectrum (FIG. 14D). Thus in the example of FIG. 14D,
sequencing analysis is conducted by filling collision
cell 304 with buffer gas and operating QMF 302 to
selectively filter out ions d and f, for example, such

25   that the resulting fragmentation spectrum includes
fragmentation spectra of ions a, b, c, e and g. The
sequencing analysis is repeated by controlling QMF 302
to selectively filter out ions c and e such that the
resulting fragmentation spectrum includes fragmentation

30   spectra of at least ions d and f. In general, the

instrument 300 must be taken through an ion
generation/resulting spectrum sequence Z + 1 times for
any sample, wherein Z is the maximum number of ions
overlapping in drift time and the "1" accounts for the
5   initial operation of instrument 300 in order to produce
the spectrum 456 of FIG. 14D. In the example
illustrated in FIGS. 14, 15 and 16, instrument 300 must
accordingly be taken through the ion
generation/resulting spectrum sequence three times since
10 the maximum number of ions overlapping in drift time is
two (e.g., two ions c and d overlap in drift time and,
two ions f and e overlap in drift time).

Referring again to FIG. 13, process 400 continues
from step 412 and step 414 where process 400 is directed
15 to the subprocess flagged with the current value of A.
In the first time through process 400, A=1 so process
400 jumps to step 416. Thereafter at step 418, the
collision cell 304 is filled with buffer gas from buffer
gas source 306 (or buffer gas source 46). As with step
20 404, step 418 may be executed manually or under the
control of computer 310. In either case, process 420
advances from step 418 to step 420 where a determination
is made as to whether there exists any overlap in ion
packet drift times. Step 420 is preferably carried out
25 by manually observing spectrum 456 (FIG. 14D), although
the present invention contemplates that step 420 may be
automated in accordance with known techniques and
therefore executed by computer 310. In either case, if
no overlap in ion drift times are present in the
30 spectrum resulting at step 412, steps 408-412 are
repeated and a spectrum of fragmented parent and

daughter ions results, wherein the spectrum of
fragmented parent and daughter ions may be analyzed
further for sequencing purposes. If, however, ion drift
time overlap is observed in the first execution of step

5 412, process 400 continues from step 420 at step 422
where QMF 302 is configured to selectively filter out
desired m/z values based on the observed overlapping
drift times. Thereafter, the process counter A is
incremented and steps 408-412 are repeated.

10 Referring now to FIGS. 15A-15D, step 422 and a
second pass through steps 408, 410 and 412 are
illustrated. The ion gate signal 450 and TOFMS signals
452 are identical to those shown in FIGS 14A and 14B,
but the QMF signal 458 includes an activation pulse 458_1

15 during a time period encompassing the drift times of
ions c and d, and an activation pulse 458_2 encompassing
the drift times of ions e and f. It is to be understood
that activation pulses 458_1 and 458_2 are not meant to
represent a single-signal activation of QMF 302 (i.e.,

20 "triggering"), but are instead meant to represent the
activation times of QMS 302 relative to known ion drift
times, wherein computer 302 is operable during each of
these activation times to control the voltage sources
320_1 and 320_2 (FIG. 11), as described hereinabove, to

25 thereby pass only ions having a desired m/z value and to
filter out ions having any other m/z value. In the
example spectrum illustrated in FIG. 15D, computer 310
is operable to control QMF 302 during activation time
458_1 to pass only ions having m/z values equal to that of

30 ion c so that ion d is effectively filtered out.
Similarly, computer 310 is operable to control QMF 302

during activation time 458₂ to pass only ions having m/z values equal to that of ion e so that ion f is effectively filtered out. In one preferred embodiment of process 400, computer 310 is operable at all other

5   times in an all-pass mode to thereby pass therethrough ions having any m/z value. In an alternate embodiment, computer 310 may be operable to sequentially control QMF 302 during time periods corresponding to the drift times of each of the ions, wherein computer 310 is operable

10   during such time periods to pass only ions having m/z values equal to those of interest. Thus, for the example spectrum 460 illustrated FIG. 15D, computer 310 may alternatively be operable to activate QMF 302 during the drift time of ion a to pass only ions having m/z

15   values equal to that of ion a, to activate QMF 302 during the drift time of ion b to thereby pass only ions having m/z values equal to that of ion b, to activate QMF 302 during the drift time of ions c and d to pass only ions having m/z values equal to that of ion c, etc.

20   In either case, the spectrum 460 of FIG. 15D results, wherein the flight times of each of the parent and daughter ions resulting from the fragmentation of ions a, b, c, e and g in collision cell 304 are clearly resolved. From these flight times, the m/z values of

25   each of the fragmented ions may be determined in accordance with known techniques.

          Referring again to FIG. 13, process 400 advances from a second execution of step 412 to step 414 where process 400 is directed to a process section flagged by

30   the most recent value of the counting variable A. In this case, A=2 so process 400 is directed to step 426.

Thereafter at step 428, a determination is made as to whether any ion packets exist that have not yet been accounted for in the spectrum 460 of FIG. 15D. In one preferred embodiment, step 428 is conducted manually via

5   examination of spectra 456 and 460, although the present invention contemplates that step 428 may alternatively be automated in a known manner and accordingly be executed by computer 310. In any case, if it is determined at step 428 that no ion packets are

10   unaccounted for, process 400 advances to step 432 where process 400 is terminated. If, on the other hand, it is determined at step 428 that there exists at least one ion packet that has not yet been accounted for in spectrum 460, process 400 advances to step 430 where QMF

15   302 is configured to selectively filter out desired m/z values based on the observed overlapping drift times. Thereafter, steps 408-412 are again repeated.

Referring now to FIGS. 16A-16D, step 430 and a third pass through steps 408, 410 and 412 are

20   illustrated. The ion gate signal 450 and TOFMS signals 452 are identical to those shown in FIGS 14A and 14B, but the QMF signal 462 includes an activation pulse 462_1 during a time period encompassing the drift times of ions c and d, and an activation pulse 462_2 encompassing

25   the drift times of ions e and f. Again, it is to be understood that activation pulses 462_1 and 462_2 are not meant to represent a single-signal activation of QMF 302 (i.e., "triggering"), but are instead meant to represent the activation times of QMS 302 relative to known ion

30   drift times, wherein computer 302 is operable during each of these activation times to control the voltage

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sources 320_1 and 320_2 (FIG. 11), as described
hereinabove, to thereby pass only ions having a desired
m/z value and to filter out ions having any other m/z
value. In the example spectrum illustrated in FIG. 16D,

5   computer 310 is operable to control QMF 302 during
activation time 462_1 to pass only ions having m/z values
equal to that of ion d so that ion c is effectively
filtered out. Similarly, computer 310 is operable to
control QMF 302 during activation time 462_2 to pass only

10   ions having m/z values equal to that of ion f so that
ion e is effectively filtered out. In one preferred
embodiment of process 400, computer 310 is operable at
all other times in a no-pass mode to thereby inhibit
passage therethrough of ions having any m/z value. In

15   an alternate embodiment, computer 310 may be operable to
sequentially control QMF 302 during time periods
corresponding to the drift times of each of the ions,
wherein computer 310 is operable during such time
periods to pass only ions having m/z values equal to

20   those of interest. Thus, for the example spectrum 464
illustrated FIG. 16D, computer 310 may additionally be
operable to activate QMF 302 during the drift times of
ions a, b and g to pass only ions having m/z values
equal to those of ions a, b and g respectively. This

25   will result in redundant flight time information for
parent/daughter ions of a, b and g, but such operation
serves as an accuracy check on the data obtained from
spectrum 464. In the first case, the spectrum 464 of
FIG. 16D results, wherein the flight times of each of

30   the parent and daughter ions resulting from the
fragmentation of ions d and f in collision cell 304 are

clearly resolved. In the latter case, a spectrum similar to spectrum 460 of FIG. 15D results, wherein the flight times of each of the parent and daughter ions resulting from the fragmentation of ions a, b, d, f and

5   g in collision cell 304 are clearly resolved. In either case, the m/z values of each of the fragmented ions may be determined from their associated flight times in accordance with known techniques.

While the invention has been illustrated and
10   described in detail in the drawings and foregoing description, the same is to be considered as illustrative and not restrictive in character, it being understood that only the preferred embodiments have been shown and described and that all changes and
15   modifications that come within the spirit of the invention are desired to be protected. For example, referring to FIG. 17, alternative variations of the ion mobility and mass spectrometer instrument of FIG. 9 are illustrated, wherein ion trapping, ion mass filtering
20   and ion fragmentation functions may, in accordance with the present invention, be positioned in various locations with respect to the ion source 32, ion mobility instrument 34 and time-of-flight mass spectrometer 36. In a first specific example, structure
25   500 represents a quadrupole mass filter, such as QMF 302 described hereinabove, structures 502 and 504 may be omitted, and structure 506 represents a collision cell such as collision cell 304. In this embodiment, ion mass selection is performed prior to injecting ions into
30   IMS 34, and ion fragmentation is performed between IMS 34 and TOFMS 36. In a second specific example,

structure 500 represents a quadrupole mass filter, such
as QMF 302 described hereinabove, structure 502
represents an ion trap, such as ion trap 152 described
hereinabove, structure 504 is omitted and structure 506

5   represents a collision cell such as collision cell 304
described hereinabove. In this embodiment, mass
selection is performed upon ions generated by ion source
32 and the mass selected ions are collected in the ion
trap 152 prior to injection into IMS 34. Fragmentation

10   is performed in collision cell 304 as described
hereinabove. Additionally, or alternatively,
fragmentation may also be performed in ion trap 152, as
is known in the art, if ion trap 152 is supplied with a
suitable buffer gas (not shown) and/or in IMS 34 as

15   described hereinabove. In a third specific example,
structure 500 represents a quadrupole mass filter, such
as QMF 302 described hereinabove, structure 502
represents a collision cell such as collision cell 304
described hereinabove, and structures 504 and 506 are

20   omitted. In this embodiment, mass selection is
performed upon ions generated by ion source 32 and the
mass selected ions are fragmented in collision cell 304
prior to injection into IMS 34. Fragmentation may
additionally or alternatively be performed in IMS 34,

25   and/or an additional collision cell 304 may be provided
as structure 506 for further fragmenting the ions
supplied by IMS 34. In a fourth specific example,
structure 500 represents a quadrupole mass filter, such
as QMF 302 described hereinabove, structure 502

30   represents an ion trap, such as ion trap 152 described
hereinabove, structure 504 represents a collision cell,

such as collision cell 304 described hereinabove, and
structure 506 is omitted. In this embodiment, mass
selection is performed upon ions generated by ion source
32, followed by collection of the mass filtered ions

5   within ion trap 152, followed by fragmentation of the
ions collected in trap 152 either within trap 152 and/or
within collision cell 304 prior to injection of the ions
into IMS 34. Further fragmentation may be performed
within IMS 34 and/or structure 506 may define an

10   additional collision cell for further ion fragmentation
prior to injection of the ions into TOFMS 36.
Generally, it is to be understood that ion mass
selection and ion fragmentation may occur at various and
multiple locations relative to ion source 32, IMS 34 and

15   TOFMS 36. Moreover, it is to be understood that IMS 34
may be generally configured as a known gas
chromatograph, as illustrated hereinabove, or
alternatively as a known liquid chromatograph, without
detracting from the scope of the present invention.

20

What is claimed is:

1.   A method of generating ion mass spectral information, comprising the steps of:

generating a gaseous bulk of ions;

gating at least a portion of said bulk of ions into an ion mobility spectrometer to thereby separate said bulk of ions in time to form a number of ion packets each having an ion mobility associated therewith;

sequentially directing at least some of said ion packets into a mass spectrometer;

continually activating said mass spectrometer to thereby sequentially separate at least some of said ion packets in time to form a number of ion subpackets each having an ion mass associated therewith; and

processing at least some of the ion subpackets to determine mass spectral information therefrom.

2.   The method of claim 1 wherein the step of generating a gaseous bulk of ions includes generating said gaseous bulk of ions from a liquefied sample.

3.   The method of claim 2 wherein the step of generating a gaseous bulk of ions from a liquefied sample includes generating said gaseous bulk via electrospray ionization.

4.   The method of claim 1 wherein the step of generating a gaseous bulk of ions includes desorbing said gaseous bulk of ions from a surface of a sample.

5.   The method of claim 4 wherein the step of
desorbing said gaseous bulk of ions from a surface of a
sample includes generating said gaseous bulk of ions via
laser desorption ionization.

5

6.   The method of claim 1 wherein the step of
generating a gaseous bulk of ions includes the steps of:
        generating gaseous ions from a sample source;
        collecting at least some of said generated gaseous

10   ions in an ion trap;
        repeating said generating and collecting steps a
number of times to thereby form a gaseous bulk of ions
in the ion trap; and
        releasing said gaseous bulk of ions from said ion

15   trap.

7.   The method of claim 6 wherein the step of
generating gaseous ions from a sample source includes
generating said gaseous ions via electrospray

20   ionization.

8.   The method of claim 6 wherein the step of
generating gaseous ions from a sample source includes
generating said gaseous ions via laser desorption

25   ionization.

9.   The method of claim 1 wherein the step of
generating a gaseous bulk of ions includes the steps of:
        continually generating gaseous ions from a sample

30   source; and

collecting a bulk of said continually generated gaseous ions in an ion collection chamber;

and wherein the step of gating at least a portion of said bulk of ions into an ion mobility instrument
5   includes gating at least a portion of said bulk of ions from said ion collection chamber into said ion mobility instrument.

10.   The method of claim 9 wherein the step of
10   generating gaseous ions from a sample source includes generating said gaseous ions via electrospray ionization.

11.   The method of claim 9 wherein the step of
15   generating gaseous ions from a sample source includes generating said gaseous ions via laser desorption ionization.

12.   The method of claim 1 further including the
20   step of sequentially fragmenting said ion packets into daughter ions prior to sequentially directing at least some of said ion packets into a mass spectrometer.

13.   The method of claim 1 further including the
25   step of selectively filtering said ion packets to thereby sequentially provide ion packets having only desired mass-to-charge ratios prior to sequentially directing at least some of said ion packets into a mass spectrometer.
30

14. The method of claim 13 further including the
step of sequentially fragmenting said ion packets having
only desired mass-to-charge ratios into daughter ions
prior to sequentially directing at least some of said
5   ion packets into a mass spectrometer.

15. The method of claim 1 further including the
step of fragmenting said gaseous bulk of ions into
daughter ions prior to said gating step.
10

16. The method of claim 1 further including the
step of selectively filtering said gaseous bulk of ions
to thereby provide a gaseous bulk of ions having only
desired mass-to-charge ratios prior to said gating step.
15

17. The method of claim 16 further including the
step of fragmenting said gaseous bulk of ions having
only desired mass-to-charge ratios into daughter ions
prior to said gating step.
20

18. The method of claim 17 further including the
step of sequentially fragmenting said ion packets into
daughter ions prior to sequentially directing at least
some of said ion packets into a mass spectrometer.
25

19. Apparatus for generating mass spectral
information from a sample source, comprising:
    means for generating a gaseous bulk of ions from a
sample source;
30   an ion mobility spectrometer (IMS) having an ion
inlet coupled to said means for generating a gaseous

bulk of ions and an ion outlet, said IMS operable to
separate ions in time as a function of ion mobility;

    a mass spectrometer (MS) having an ion acceleration
region coupled to said ion outlet of said IMS and an ion

5   detector, said MS operable to separate ions in time as a
function of ion mass; and

    a computer operable to gate at least a portion of
said gaseous bulk of ions into said ion inlet of said
IMS and to continually pulse said ion acceleration

10   region of said MS to thereby sequentially direct ions
toward said ion detector.

    20.   The apparatus of claim 19 wherein said ion
detector is operable to produce detector output signals

15   indicative of detection of ions thereat;
    and wherein said computer is responsive to said
detector output signals to produce corresponding
spectral information of ions separated both in ion
mobility and in ion mass.

20

    21.   The apparatus of claim 19 wherein said means
for generating a bulk of gaseous ions is responsive to a
gate signal to generate said gaseous bulk of ions;
    and wherein said computer is operable to produce

25   said gate signal to thereby gate at least a portion of
said gaseous bulk of ions into said ion inlet of said
IMS.

    22.   The apparatus of claim 19 wherein said ion

30   inlet of said IMS defines an ion gate responsive to an
active gate signal to permit entrance of gaseous ions

into said ion inlet of said IMS and to otherwise inhibit entrance of gaseous ions into said ion inlet of said IMS;

5   and wherein said computer is operable to control said gate signal, said control computer activating said gate signal for a programmable time period to thereby gate at least a portion of said gaseous bulk of ions into said ion inlet of said IMS.

10   23. The apparatus of claim 19 further including a collision cell disposed between said ion outlet of said IMS and said acceleration region of said MS, said collision cell having a buffer gas therein operable to fragment parent ions provided at said outlet of said IMS

15   into daughter ions prior to entrance into said ion acceleration region of said MS.

24. The apparatus of claim 19 further including an ion mass filter disposed between said ion outlet of said

20   IMS and said ion acceleration region of said MS, said ion mass filter directing ions having only desired mass-to-charge ratios into said ion acceleration region of said MS.

25   25. The apparatus of claim 24 further including a collision cell disposed between said ion mass filter and said ion acceleration region of said MS, said collision cell having a buffer gas therein operable to fragment parent ions having only desired mass-to-charge ratios

30   into daughter ions prior to entrance into said ion acceleration region of said MS.

26. The apparatus of claim 19 further including a collision cell disposed between said means for generating a gaseous bulk of ions and said ion inlet of

5   said IMS, said collision cell having a buffer gas therein operable to fragment parent ions provided by said means for generating a gaseous bulk of ions into daughter ions prior to entrance into said ion inlet of said IMS.

10

27. The apparatus of claim 19 further including an ion mass filter disposed between said means for generating a gaseous bulk of ions and said ion inlet of said IMS, said ion mass filter directing ions having

15   only desired mass-to-charge ratios into said ion inlet of said IMS.

28. The apparatus of claim 27 further including a collision cell disposed between said ion mass filter and

20   said ion inlet of said IMS, said collision cell having a buffer gas therein operable to fragment parent ions provided by said ion mass filter into daughter ions prior to entrance into said ion inlet of said IMS.

25   29. The apparatus of claim 19 further including a collision cell disposed between said ion outlet of said IMS and said acceleration region of said MS, said collision cell having a buffer gas therein operable to fragment parent ions provided at said outlet of said IMS

30   into daughter ions prior to entrance into said ion acceleration region of said MS.

30.   A method of generating ion mass spectral
information, comprising the steps of:

generating a gaseous bulk of ions;

5   separating said gaseous bulk of ions in time as a
function of ion mobility;

where two or more ions overlap in ion mobility
values, filtering out ions that have all but a desired
mass-to-charge ratio;

10   sequentially separating in time the post-filtered
ions as a function of ion mass; and

processing ions separated in time as functions of
ion mobility and ion mass to determine ion mass spectral
information therefrom.

15

31.   The method of claim 30 further including the
step of fragmenting post-filtered parent ions into
daughter ions prior to the step of sequentially
separating the post filtered ions as a function of ion

20   mass.

32.   The method of claim 31 wherein the step of
generating a gaseous bulk of ions includes generating
said gaseous bulk of ions via electrospray ionization.

25

33.   The method of claim 31 wherein the step of
generating a gaseous bulk of ions includes generating
said gaseous bulk of ions via laser desorption
ionization.

30

34.   The method of claim 31 wherein the step of
generating a gaseous bulk of ions includes the steps of:
      generating gaseous ions from a sample source;
      collecting at least some of said generated gaseous

5   ions in an ion trap;
      repeating said generating and collecting steps a
number of times to thereby form a gaseous bulk of ions
in the ion trap; and
      releasing said gaseous bulk of ions from said ion

10   trap.

35.   The method of claim 31 wherein the step of
generating a gaseous bulk of ions includes the steps of:
      continually generating gaseous ions from a sample

15   source; and
      collecting a bulk of said continually generated
gaseous ions in an ion collection chamber; and
      releasing at least a portion of said continually
generated gaseous ions from said ion collection chamber.

20

36.   The method of claim 35 wherein the step of
generating gaseous ions from a sample source includes
generating said gaseous ions via electrospray
ionization.

25

37.   The method of claim 35 wherein the step of
generating gaseous ions from a sample source includes
generating said gaseous ions via laser desorption
ionization.

30

38. Apparatus for generating mass spectral
information from a sample source, comprising:

means for generating a gaseous bulk of ions from a
sample source;

5   an ion mobility spectrometer (IMS) having an ion
inlet coupled to said means for generating a gaseous
bulk of ions and an ion outlet, said IMS operable to
separate ions in time as a function of ion mobility;

an ion filter having a filter inlet coupled to said

10 ion outlet of said IMS and a filter outlet, said ion
filter operable to sequentially pass therethrough only
ions having desired mass-to-charge ratios; and

a mass spectrometer (MS) having an ion acceleration
region coupled to said filter outlet and an ion

15 detector, said MS operable to sequentially separate in
time ions provided thereto by said ion filter as a
function of ion mass.

39. The apparatus of claim 38 further including a

20 computer operable to gate at least a portion of said
gaseous bulk of ions into said ion inlet of said IMS and
to continually pulse said ion acceleration region of
said MS.

25   40. The apparatus of claim 39 wherein said
computer is further operable to control said ion filter
to thereby permit passage therethrough of ions having
only desired mass-to-charge ratios.

41. The apparatus of claim 40 wherein said means for generating a bulk of gaseous ions is responsive to a gate signal to generate said gaseous bulk of ions;

5   and wherein said computer is operable to produce said gate signal to thereby gate at least a portion of said gaseous bulk of ions into said ion inlet of said IMS.

42. The apparatus of claim 41 wherein said ion
10   inlet of said IMS defines an ion gate responsive to an active gate signal to permit entrance of gaseous ions into said ion inlet of said IMS and to otherwise inhibit entrance of gaseous ions into said ion inlet of said IMS;

15   and wherein said computer is operable to control said gate signal, said control computer activating said gate signal for a programmable time period to thereby gate at least a portion of said gaseous bulk of ions into said ion inlet of said IMS.

20

43. The apparatus of claim 38 further including a collision cell disposed between said ion filter outlet and said acceleration region of said MS, said collision cell having a buffer gas therein operable to fragment
25   ions provided at said ion filter outlet into parent and daughter ions prior to entrance into said ion acceleration region of said MS.

44. A method of generating ion mass spectral
30   information, comprising the steps of:
       generating a gaseous bulk of ions;

separating said gaseous bulk of ions in time as a
function of ion mobility;

sequentially separating in time as a function of
ion mass each of the ions separated in time as a

5   function of ion mobility;

processing ions separated in time as functions of
ion mobility and ion mass to determine ion mass spectral
information therefrom;

repeating the generating and separating steps

10   followed by the step of sequentially fragmenting into
daughter ions each of the ions separated in time as a
function of ion mobility, followed by the sequentially
separating and processing steps only if the initial
processing step indicates that no two or more ions

15   overlap in mobility values.

45.   The method of claim 44 further including the
step of filtering out ions that have all but a desired
mass-to-charge ratio, followed by repeating the

20   generating and separating steps, followed by the step of
sequentially fragmenting into daughter ions each of the
ions separated in time as a function of ion mobility,
followed by the sequentially separating and processing
steps only if the previous processing step indicates

25   that two or more ions overlap in mobility values.

46.   The method of claim 45 further including the
step of repeating the filtering step until all ions
overlapping in ion mobility values have been processed.

30

47.   The method of claim 46 wherein the step of
generating a gaseous bulk of ions includes generating
said gaseous bulk of ions via electrospray ionization.

5   48.   The method of claim 46 wherein the step of
generating a gaseous bulk of ions includes generating
said gaseous bulk of ions via laser desorption
ionization.

10   49.   The method of claim 46 wherein the step of
generating a gaseous bulk of ions includes the steps of:
          generating gaseous ions from a sample source;
          collecting at least some of said generated gaseous
ions in an ion trap;
15   repeating said generating and collecting steps a
number of times to thereby form a gaseous bulk of ions
in the ion trap; and
          releasing said gaseous bulk of ions from said ion
trap.
20

          50.   The method of claim 46 wherein the step of
generating a gaseous bulk of ions includes the steps of:
          continually generating gaseous ions from a sample
source; and
25   collecting a bulk of said continually generated
gaseous ions in an ion collection chamber; and
          releasing at least a portion of said continually
generated gaseous ions from said ion collection chamber.

30   51.   The method of claim 50 wherein the step of
generating gaseous ions from a sample source includes

generating said gaseous ions via electrospray
ionization.

52. The method of claim 50 wherein the step of
generating gaseous ions from a sample source includes
generating said gaseous ions via laser desorption
ionization.

TIME OF FLIGHT (microseconds)
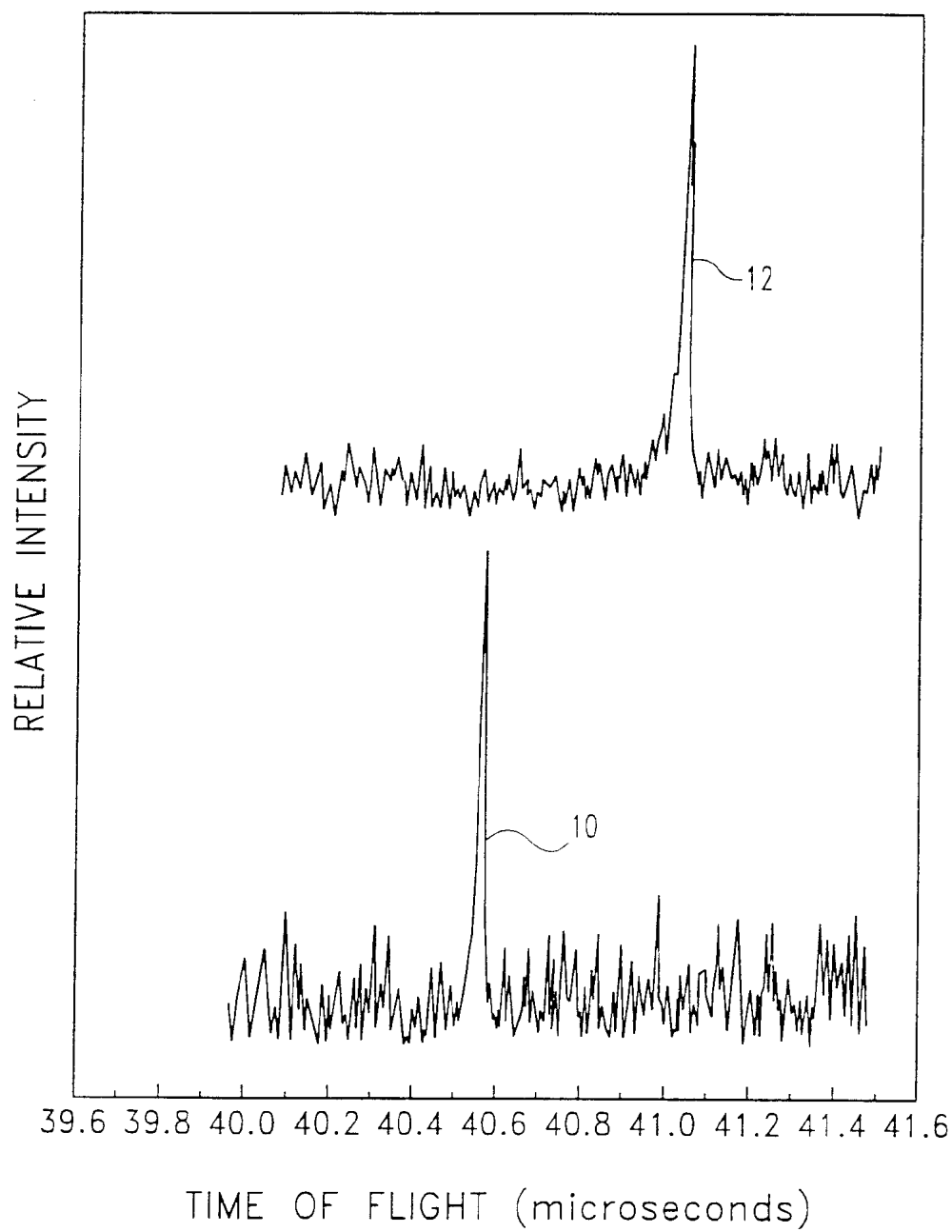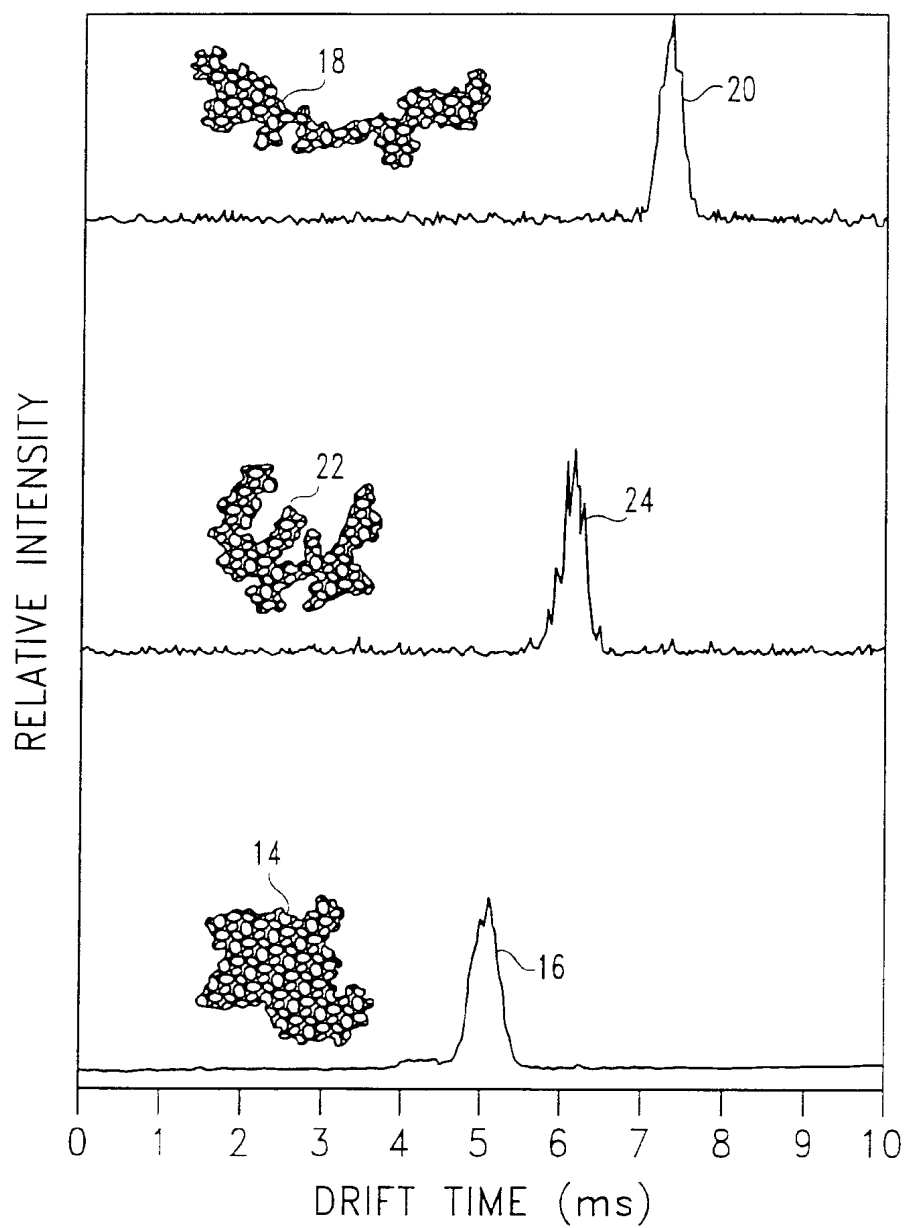
Fig. 1

(PRIOR ART)

Fig. 2

(PRIOR ART)
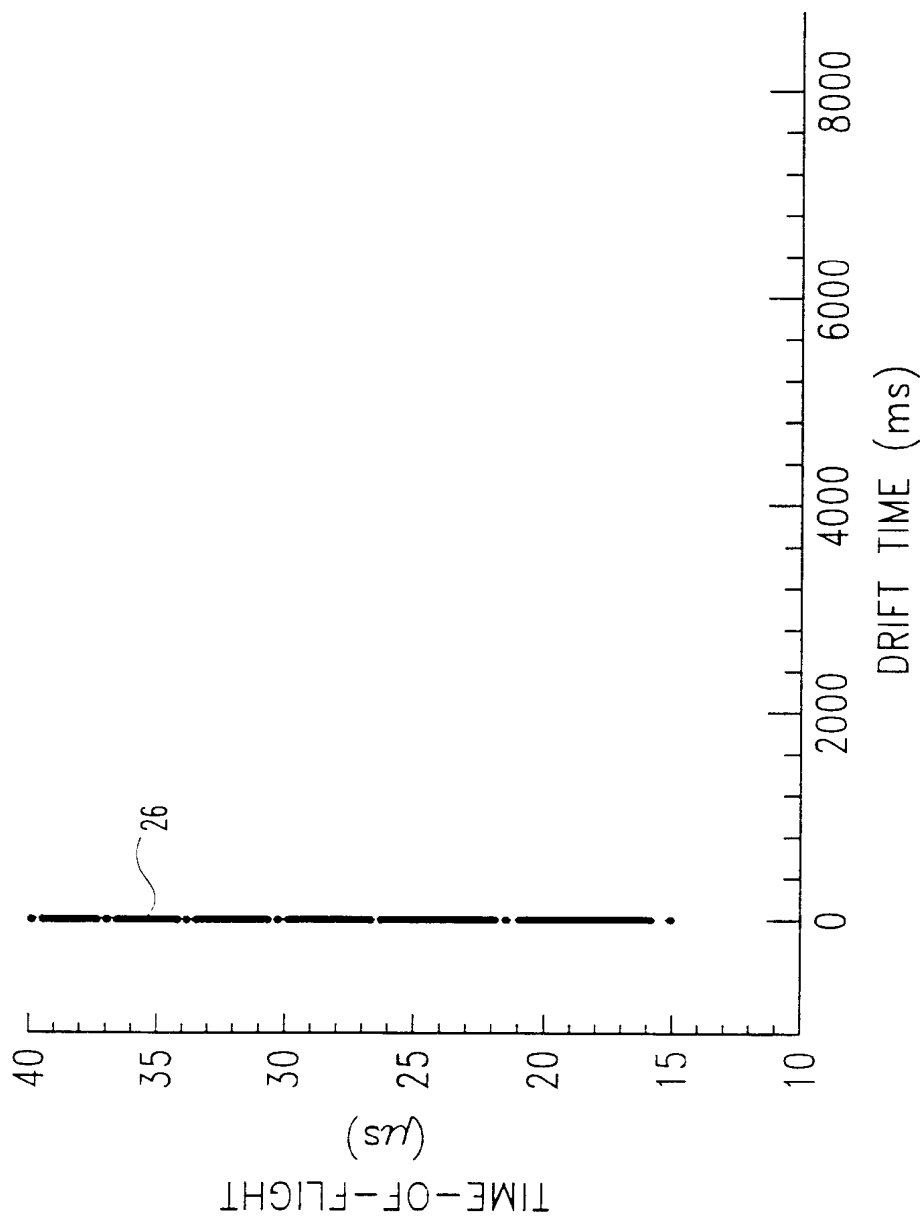
Fig. 3

Fig. 4

Fig. 5

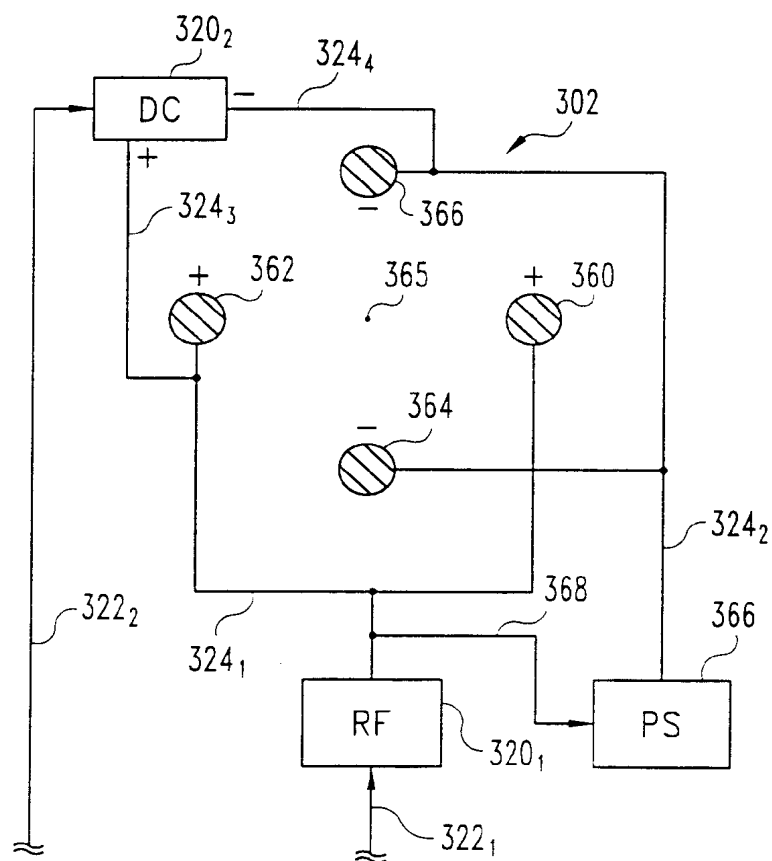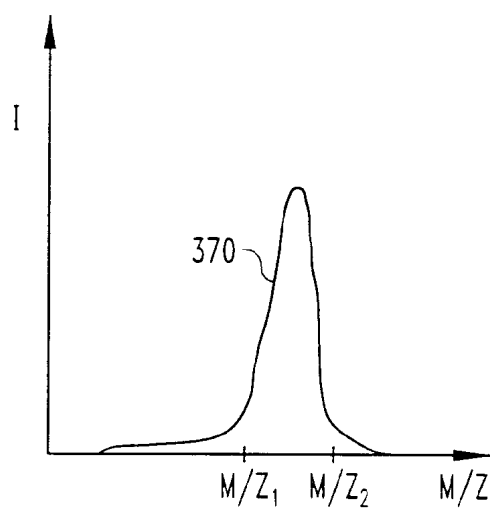Fig. 6

Fig. 7A

Fig. 7C
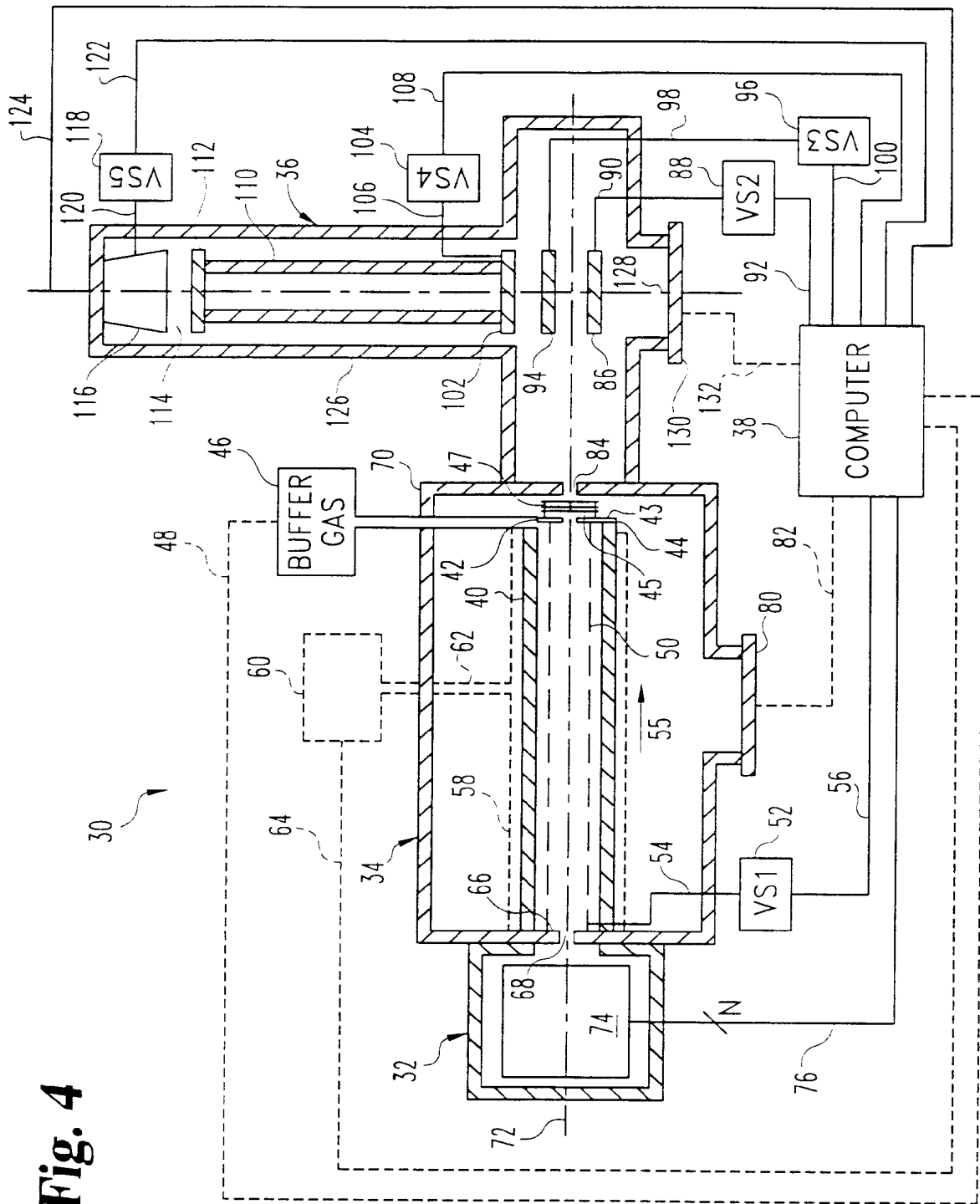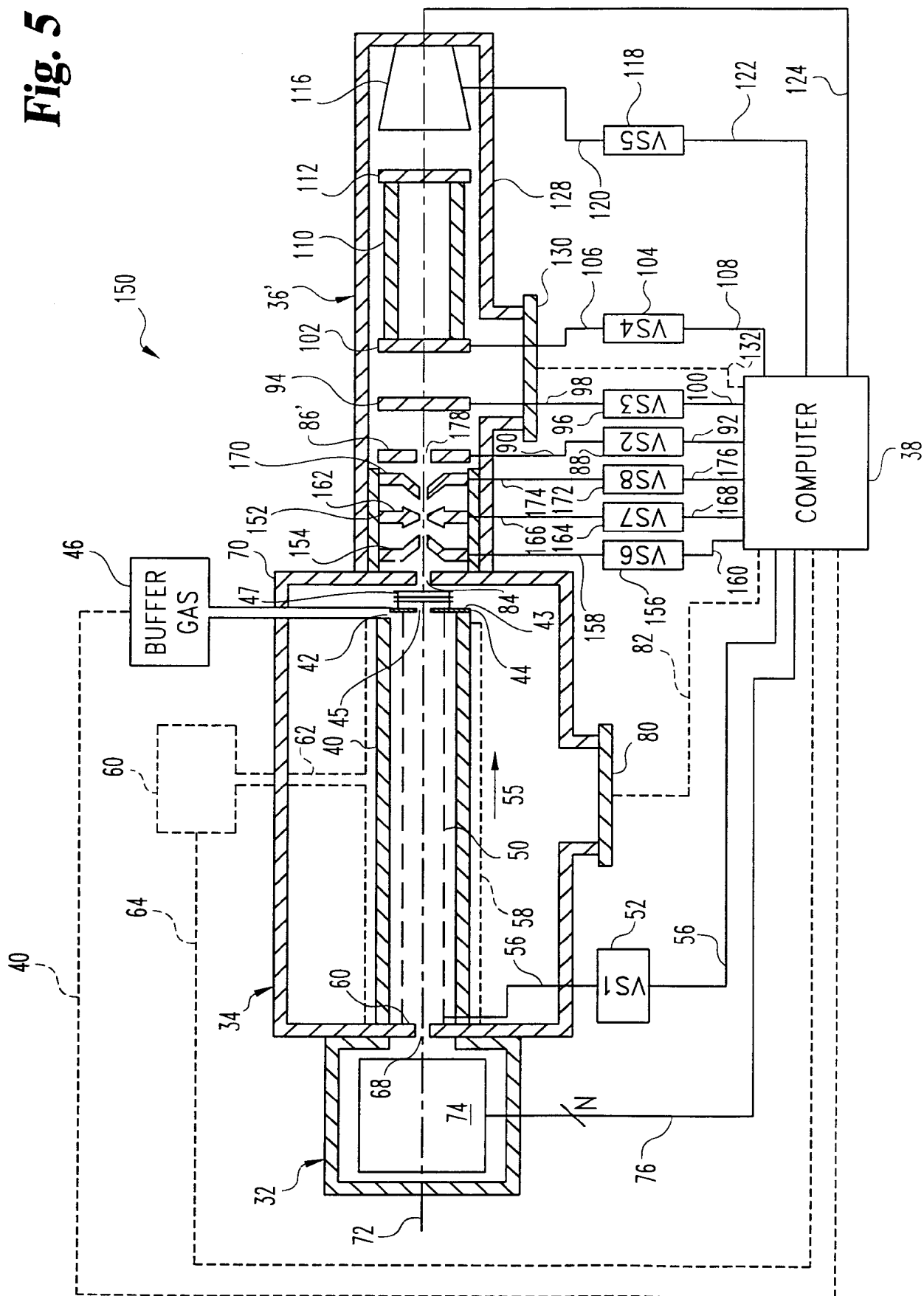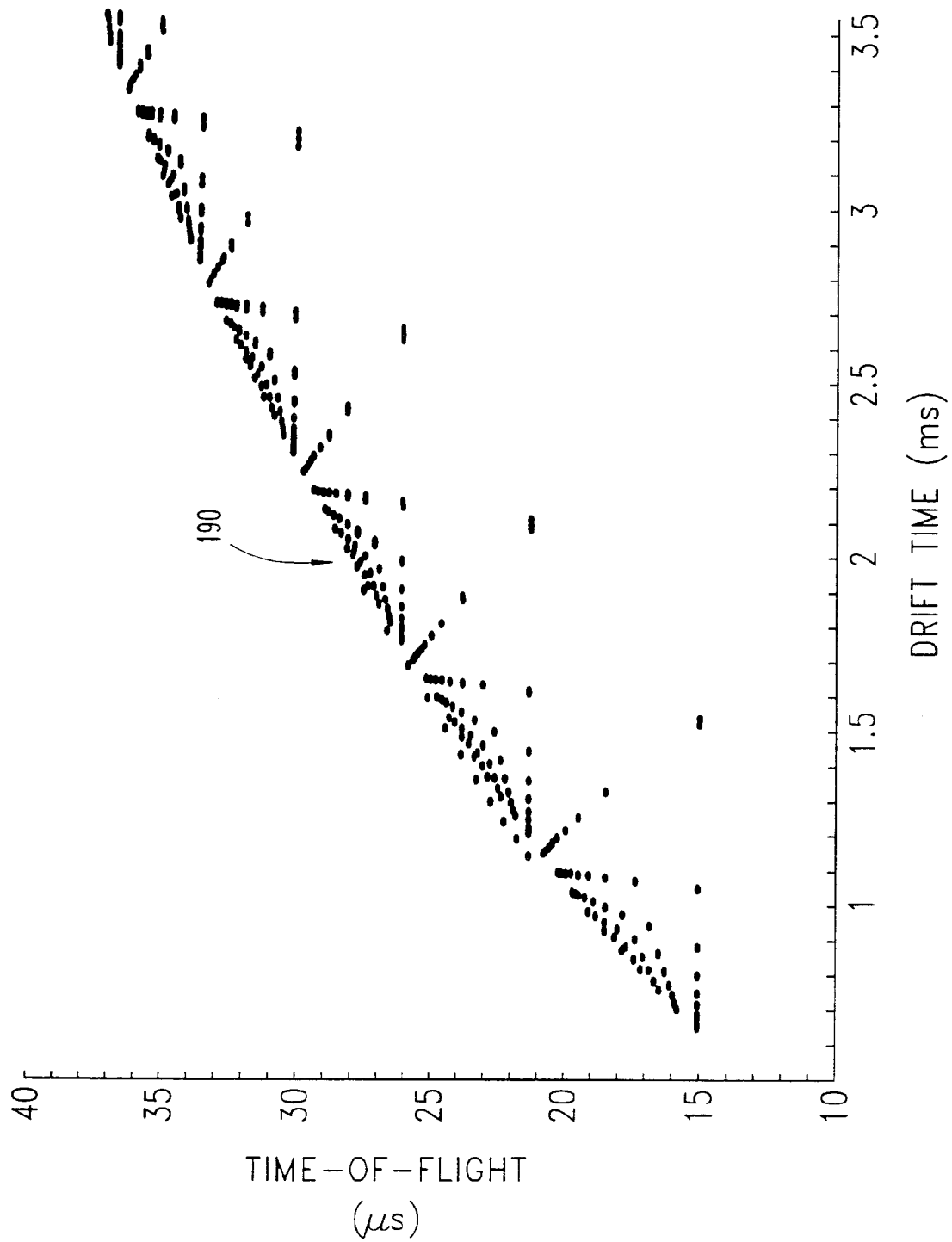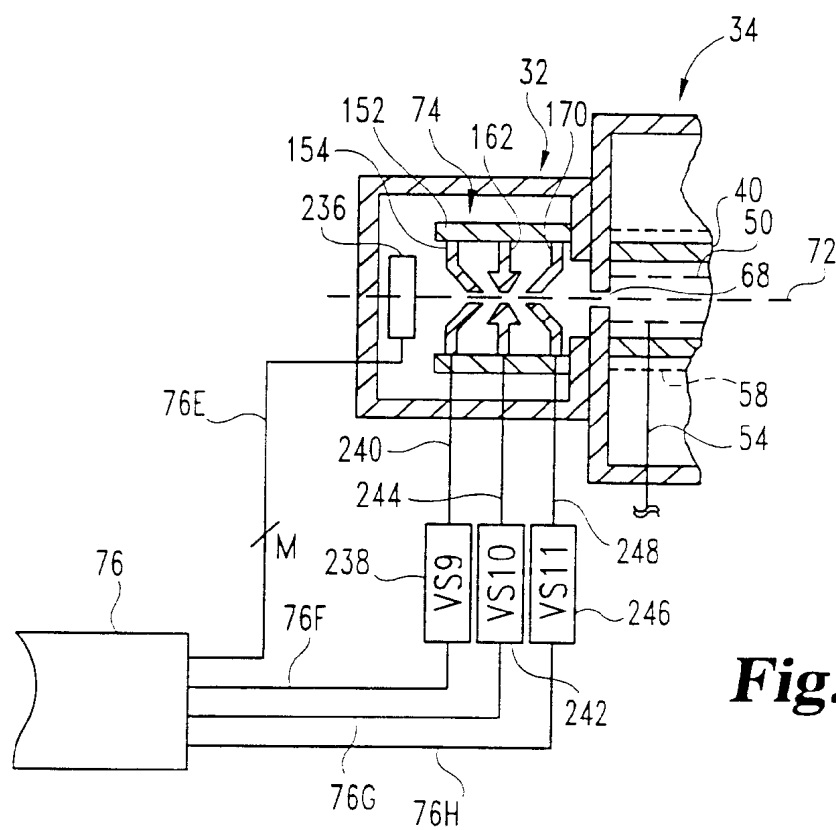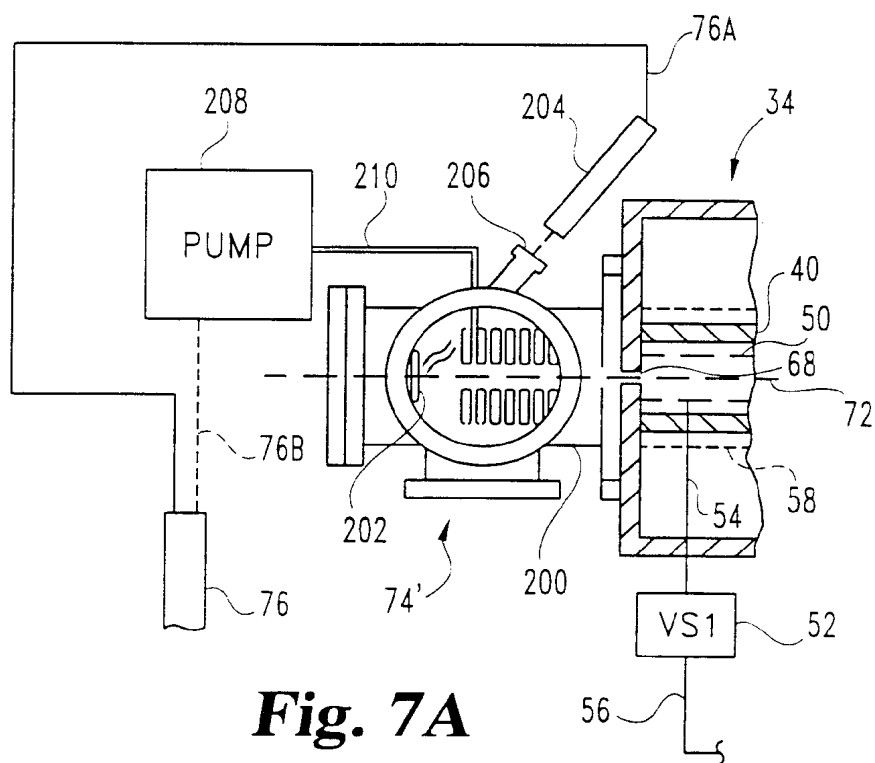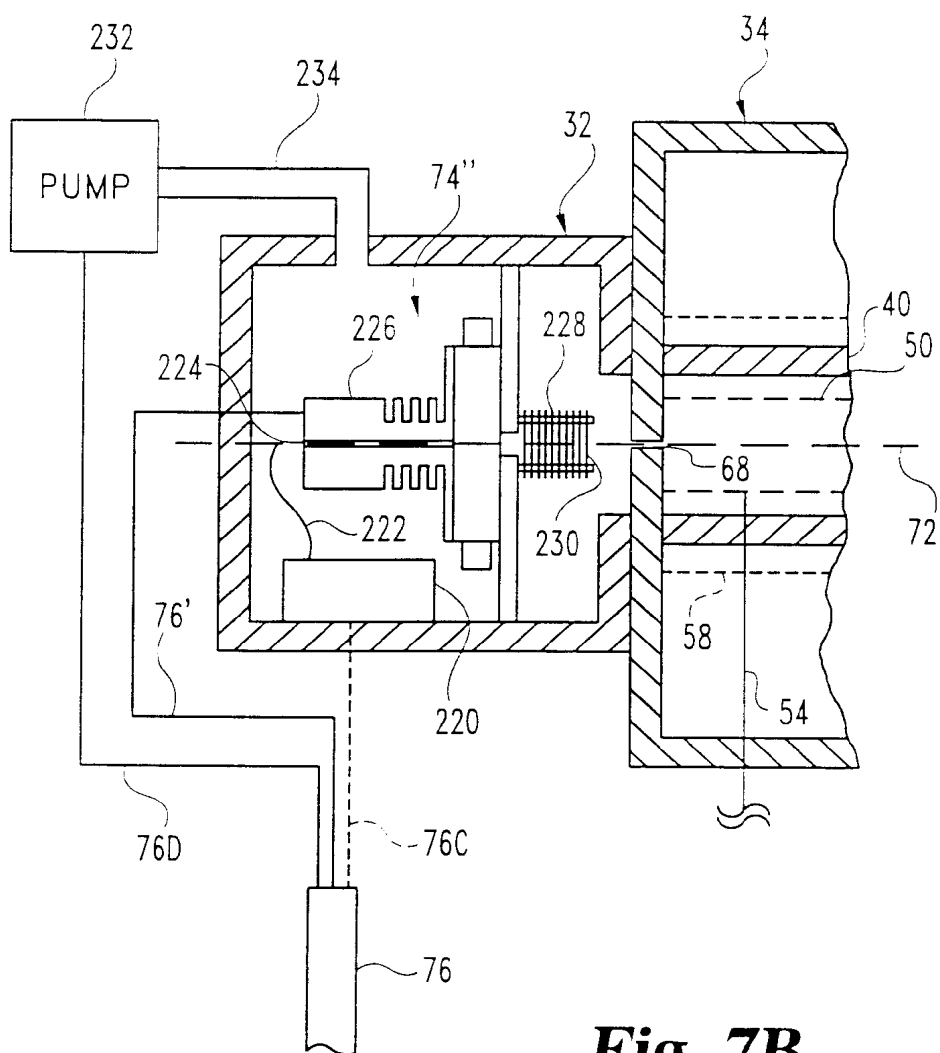
Fig. 7B

Fig. 8B

Fig. 8A

Fig. 9
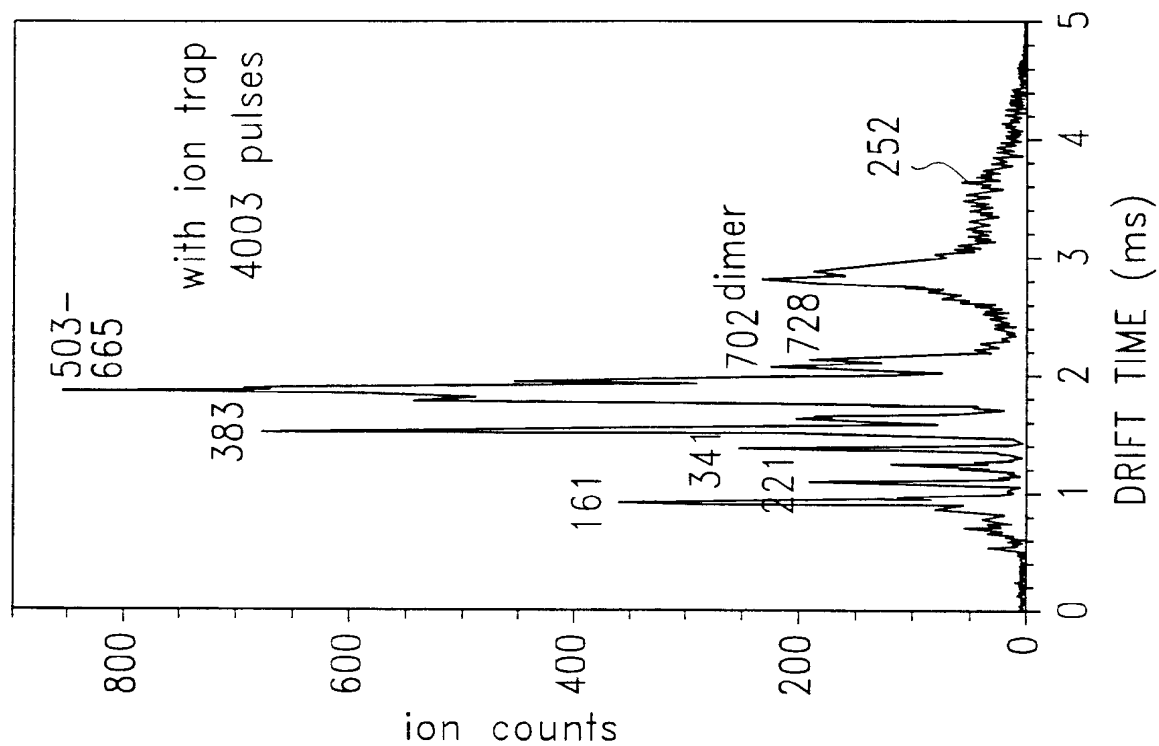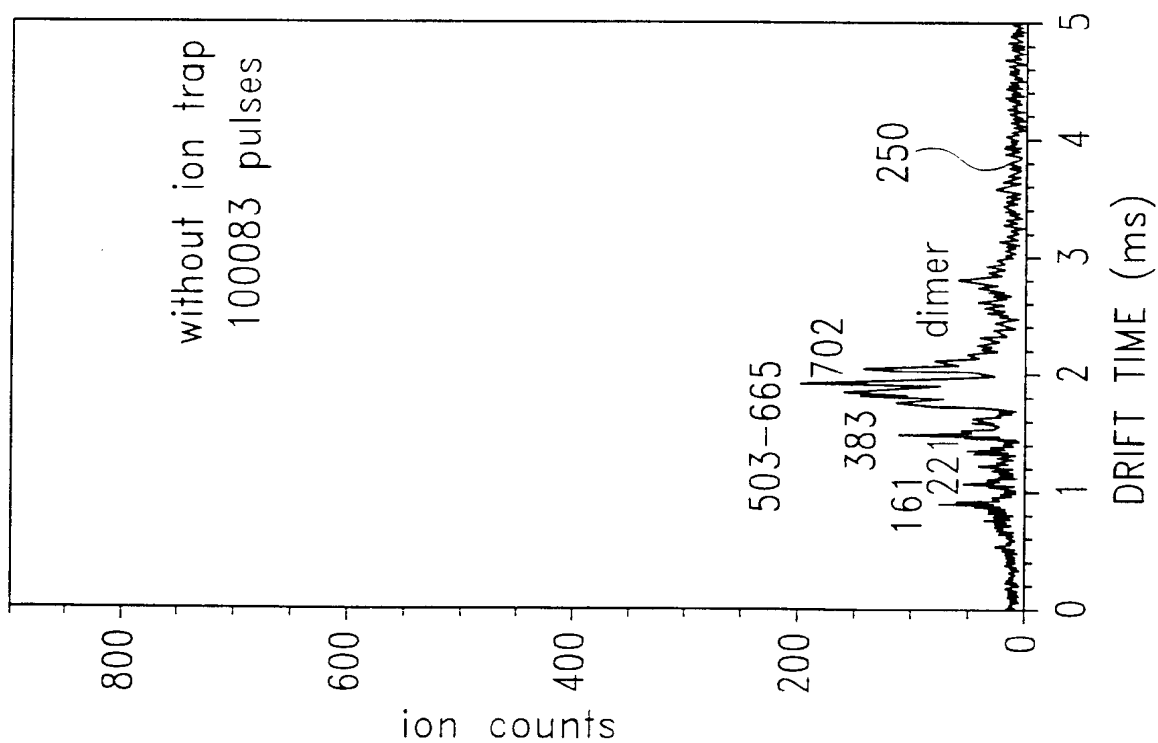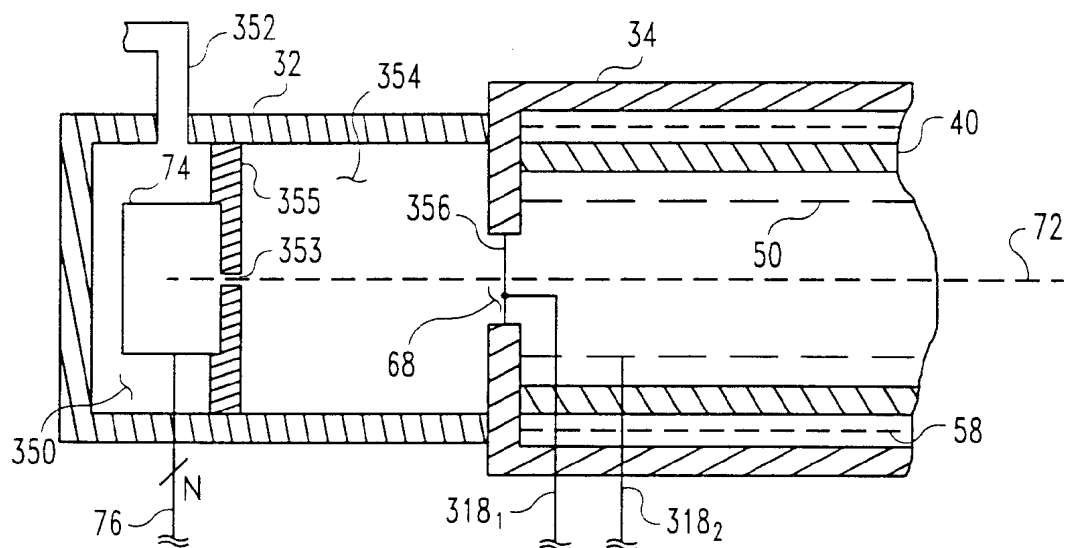
Fig. 10

Fig. 11

Fig. 12

Fig. 13
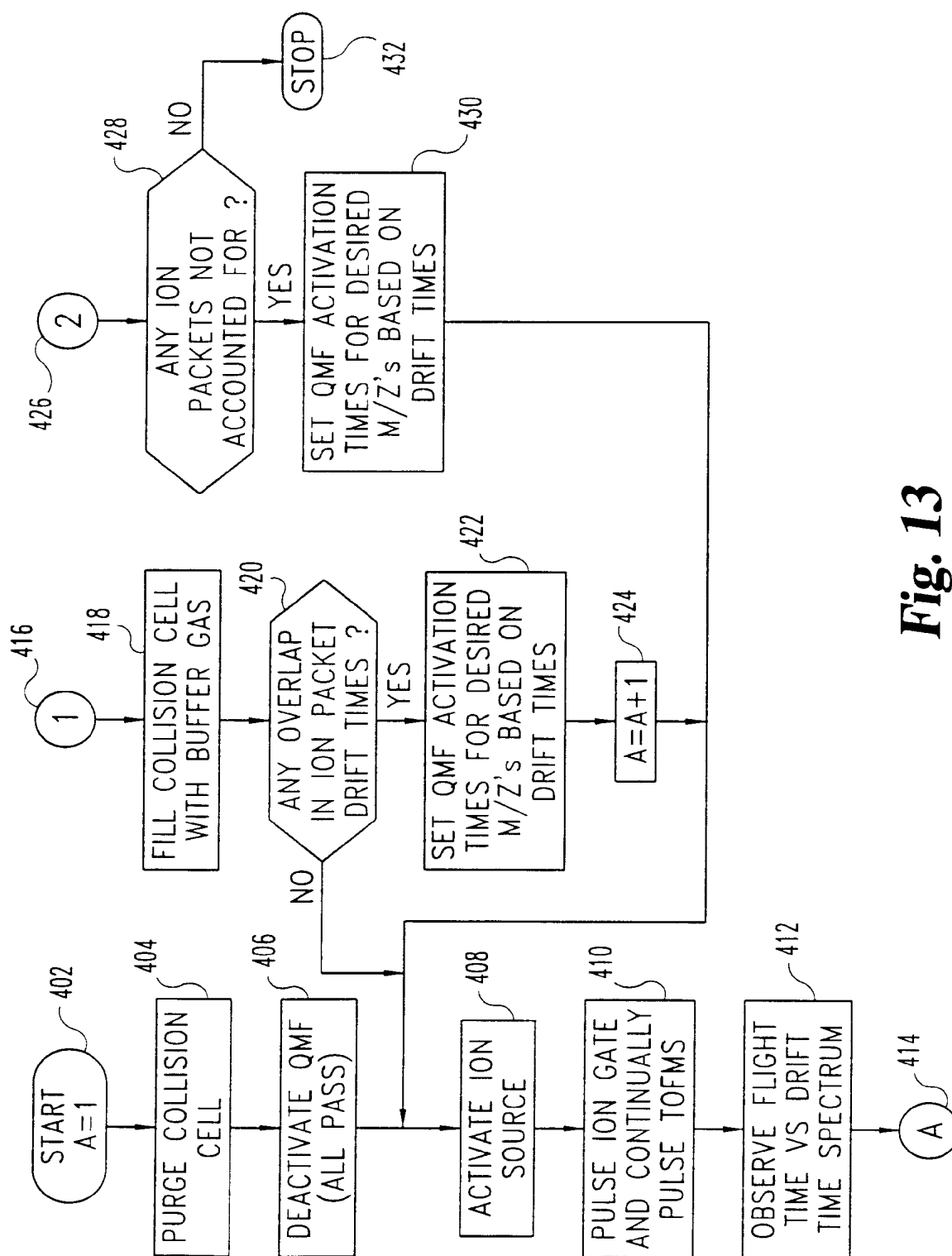
Fig. 14A

Fig. 14B

Fig. 14C

Fig. 14D

IG

450

TOFMS

452

QMF

454

FLIGHT
TIME
(μs)

9
8
7
6
5
4
3
2
1
0

● a
● b
● c
● d
● e
● f
● g

456

0   1   2   3   4   5   6   7   8
DRIFT TIME (ms)

Fig. 15A

Fig. 15B

Fig. 15C
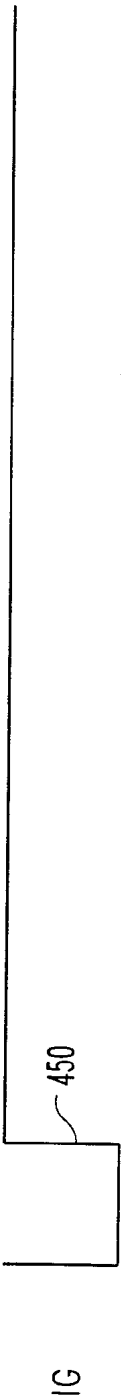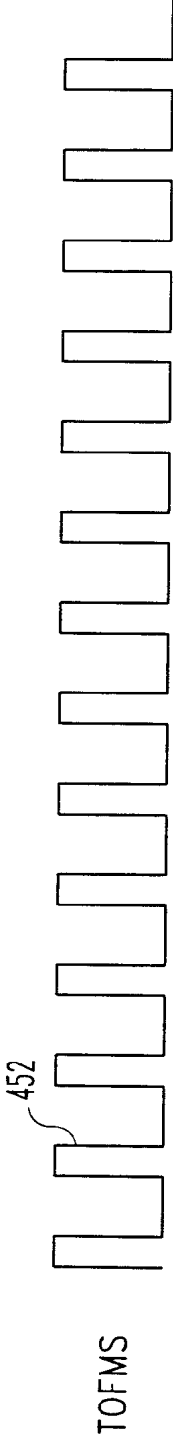
Fig. 15D
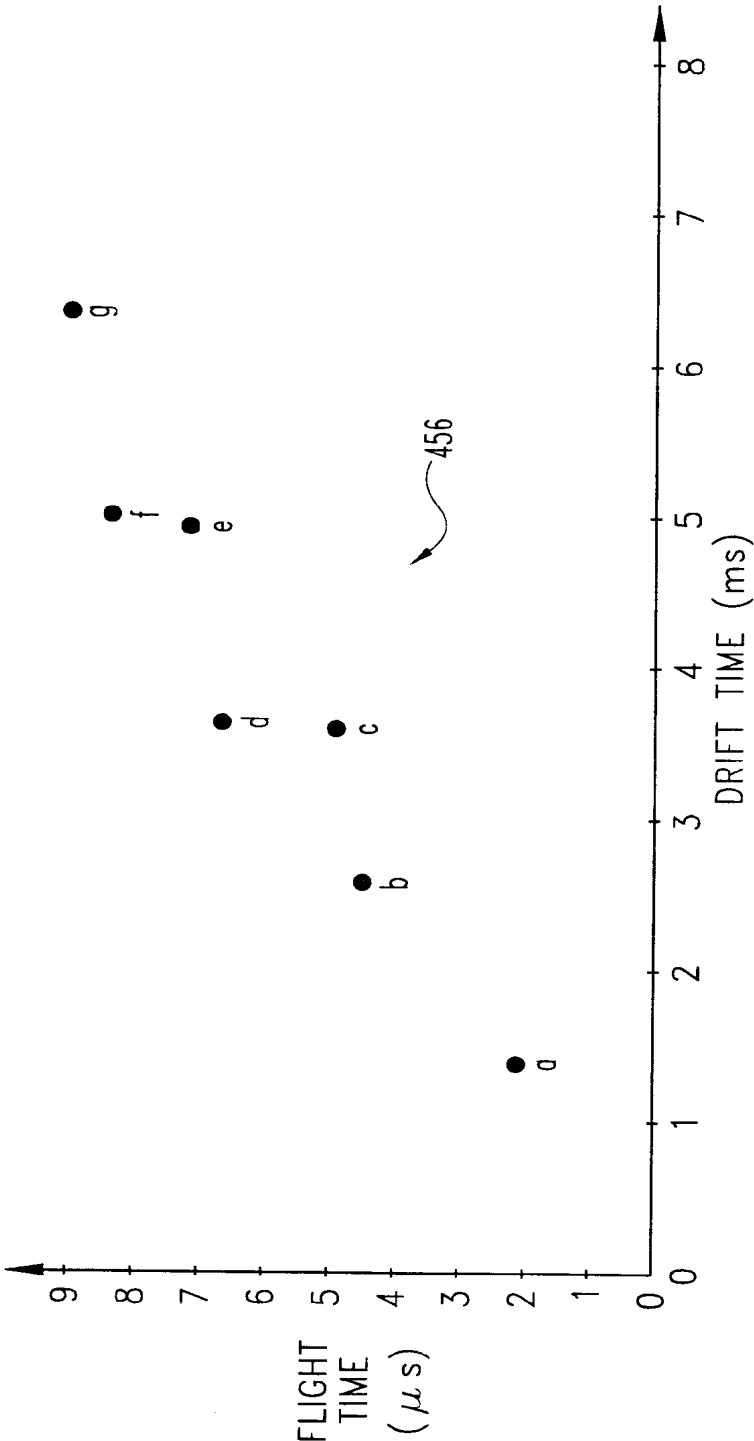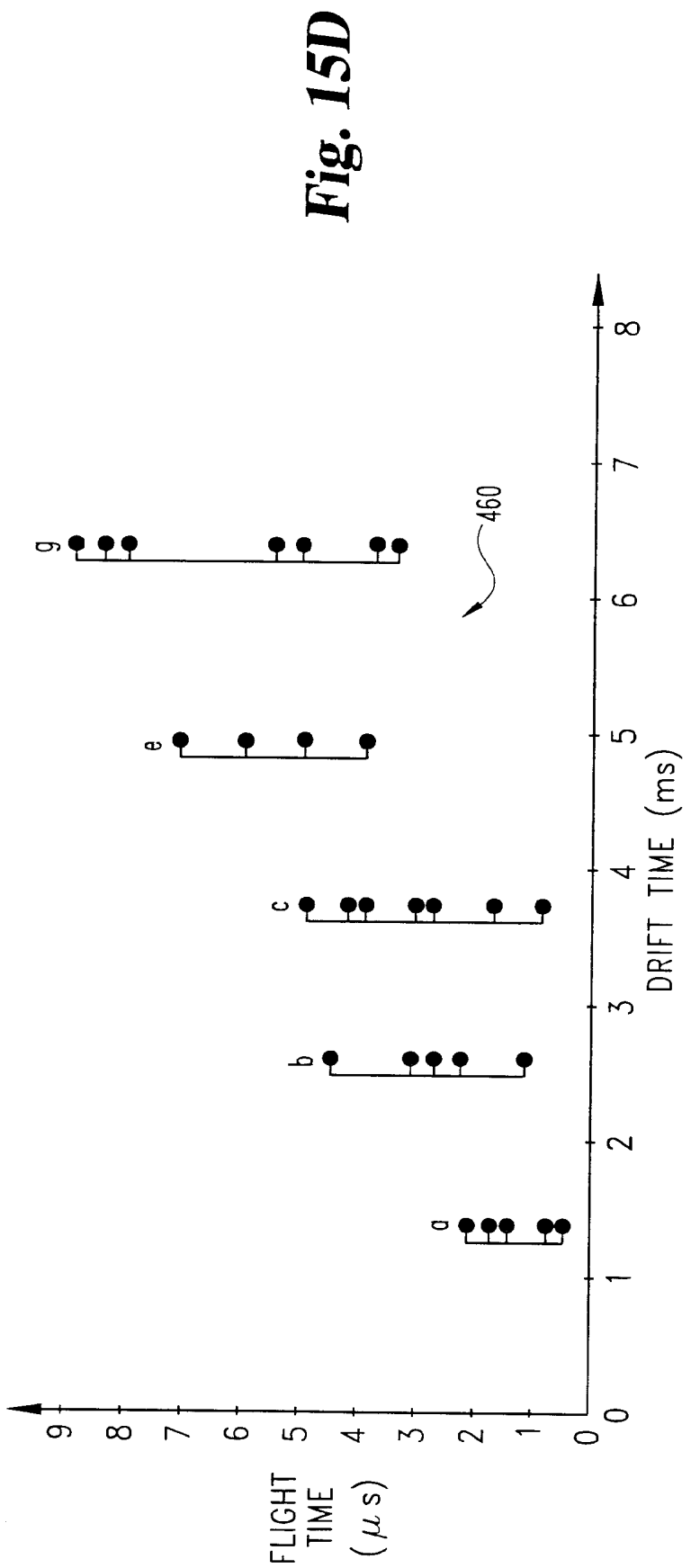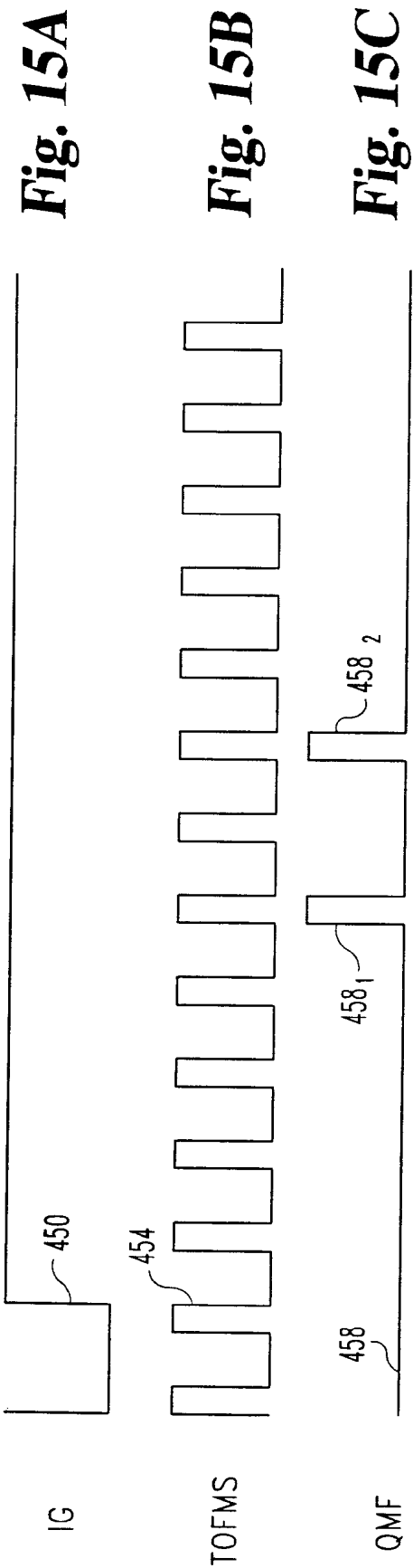
Fig. 16A

Fig. 16B

Fig. 16C

Fig. 16D

Fig. 17